Academic Sciences International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 4, Suppl 3, 2012

Research Article

ANTIMICROBIAL ACTIVITY OF HEARTWOOD OF *TECOMA STANS.*

DR.A.KOTTAI MUTHU[2], LAXMIKANT.B.BORSE[1*], DR.A.THANGATRIPATHI[3], SANDHYA.L.BORSE[4]

[1]Dept. of Pharmacology, Karpagam University, Coimbatore, [2]Department of Pharmacy, Annamalai University, Annamalai Nagar-608 002, [3]S.B.College of Pharmacy, Sivakasi, [4]P.S.G.V.P.Mandal's College of Pharmacy, Shahada-425409, India.
Email: laxmikantborse15@rediffmail.com

Received: 19 Jan 2012, Revised and Accepted: 09 Mar 2012

ABSTRACT

The aim of the present study was to evaluate the antimicrobial activity of various extracts from heartwood of *Tecoma stans*. The different solvent extracts of *Tecoma stans* effective against tested bacteria *(Pseudomonas fluorescens*, *Clavibacter michiganensis* sub sp. *michiganensis*, *Xanthomonas axanopodis* pv. *malvacearum*, *Staphylococcus aureus, E. coli*, *Pseudomonas aeruginosa* and *Klebsiella pneumonia*) and fungi (all species of *Aspergillus* and *Alternaria*). In phytochemical screening showed that the different solvent extracts of *Tecoma stans*, the tannin, flavonoids, phenol, alkaloids, steroids, anthraquinones and saponins were present in all solvent extracts. The Strong antimicrobial activities were observed in the ethanolic and methanolic extracts of *T. stans* than that of water extract. These findings provide scientific evidence to support traditional medicinal uses and indicate a promising potential for the development of an antimicrobial agent from *T. stans* plant. This medicinal plant by *in vitro* results appears as interesting and promising and may be effective as potential sources of novel antimicrobial drugs.

Keywords: *Tecoma stans*, Antimicrobial, Phytochemicals.

INTRODUCTION

The increase in prevalence of multiple drug resistance has showed down the development of new synthetic antimicrobial drugs and the new drug is necessary to search for new antimicrobial from alternative sources. Phytochemicals from medicinal plants showing antimicrobial activities have the potential of filling this need because of structures are different from those of the more studied and their more action may too very likely differ[1]. In this growing interest, many of the Phytochemical bioactive compounds from a medicinal plants have shown many pharmacological activities [2]. Screening of various bioactive compounds from plants has lead to the discovery of new medicinal drug which have efficient protection and treatment roles in against various diseases[3].

The rapid emergence of multiple drug resistance strains of pathogens to current antimicrobial agents has generated an urgent intensive for new antibiotics from medicinal plants. Many medicinal plants have been screened extensively for their antimicrobial potential worldwide [4]. Free radicals which have one or more unpaired electrons (superoxide, hydroxyl, peroxyl) are produced in normal or pathological cell metabolism and the compounds that can scavenge free radicals have great potential in ameliorating the diseases and pathological cells [5].

Tecoma stans (Bignoniaceae) known as yellow elder is an erect shrub or small tree. The plant has been used for a variety of purposes in herbal medicine, treating diabetes and digestive problems. Extracts from *Tecoma stans* leaves have been found to be inhibit the growth of the yeast infection. Senthilkumar et al. (2010) have reported that the extracts having antibacterial activity on human pathogenic bacteria. In present study was aimed to examine the total Phenolic content and Phytochemical analysis of ethanol, methanol and water extract of *Tecoma stans* were screened for antimicrobial properties using standard methods. The findings from this work may add to the overall value of the medicinal potential of the plant.

MATERIALS AND METHODS

Plant material

The heartwood of *Tecoma stans* were collected from the medicinal garden of the college of Pharmacy Shahada, In August and were air dried at room temperature for 4 weeks and were authenticated by Botanical survey of India, Koregaon road, Pune.

Preparation of extracts

Heartwood of *Tecoma stans* Plant were air dried at room temperature for 4 weeks to get consistent weight. The dried parts were later ground to powder. 100 g of wet and dried samples were extracted with distilled water, ethanol, and methanol separately for 2 days in water both with a shaking attachment. The extract was lyophilized under 5 µm Hg pressure and stored at -200C. The experimental were carried out using an appropriate amount of lyophilized material.

Phytochemical analysis

Phytochemical analysis was carried out for saponins, flavonoids, steroids, phenol, anthroquinone, alkaloids and tannins. Wagner's and Heger's reagents was used for alkaloid foam test for saponins, Mg- HCl and Zn-HCl for flavonoids, acetic anhydride and sulphuric acid for steroids, chloride and gelatin for tannins, ferric chloride for phenol, hexane and diluted ammonia for anthraquinones test. All these experiments were carried out for distilled water, ethanol and methanol extracts individually.

Determination of Antimicrobial Activity

Antimicrobial assay

Pseudomonas fluorescens, *Clavibacter michiganensis* sub sp. *michiganensis*, *Xanthomonas oryzae* pv. *oryzae*, *anthomonas axanopodis* pv. *malvacearum* and strains of *Staphylococcus aureus, E. coli, Pseudomonas aeruginosa* and *Klebsiella pneumonia* bacteria were obtained from stock cultures presented at -80°C at P.S.G.V.P.Mandal's Department of Microbiology, Shahada, dist-Nandubar, Maharashtra. Two Gram positive bacteria tested were *Clavibacter michiganensis* sub sp. *michiganensis*, *Staphylococcus aureus* and six Gram negative bacterias tested were *Pseudomonas fluorescens*, *Xanthomonas oryzae* pv. *oryzae*, *Xanthomonas axanopodis* pv. *malvacearum*, *E. coli*, *Pseudomonas aeruginosa* and *Klebsiella pneumonia*. All bacteria were grown on nutrient agar media. Fungi (*Aspergillus flavus, Aspergillus niger, Alternaria carthami, Alternaria helianthi, Cercospora carthami, Fusarium solani, Fusarium oxysporum, Fusarium verticilloides* and *Nigrospora oryzae* were obtained from P.S.G.V.P. Mandal's Department of Microbiology, Shahada, dist-Nandubar,Maharashtra. All fungi were grown on potato dextrose agar medium.

Paper disc method

Diameter of zone of inhibition was determined using the paper disc diffusion method as described [7]. A swab of the bacteria or fungi suspension containing 1x108 CFU/ml was spread on to Petri plates containing nutrient agar media separately. Each solvent extracts

were dissolved in each solvent to final concentration of 10mg/ml. Sterile filter paper discs (6mm in diameter) impregnated with 1mg of plant extracts were placed on culture plates separately for bacteria and fungi. The plates were incubated at 37°C for 24h. The standard chloramphenicol (10µg) for bacteria and carbendazim for fungi discs were used as positive controls. Antimicrobial activity was indicated by the presence of clear inhibition zone around the discs. The assay was repeated thrice and mean of three experiments was recorded.

RESULTS

Phytochemical analysis

The phytochemical screening showed that the different solvent extracts of *Tecoma stans*, the tannin, flavonoids, phenol, alkaloids, steroids, anthraquinones and saponins were present in all solvent extracts. The phytochemicals strongly present in the ethanol and methanol extracts. But the water extract yielded less quantity of phytochemicals (Table-1).

Table 1: Phytochemical analysis for the different solvent extracts of heartwood of *T. stans*

Extracts	Alkaloids	Flavonoids	Saponins	Tannins	Phenols	Steroids	Anthroquinones
Methanol	+++	+++	+++	+++	+++	+++	+++
Ethanol	+++	+++	+++	+++	+++	+++	+++
Water	+	+	+	+	+	+	+

+++ Strong; ++ medium; +poor Presence; -: absence, repeated the experiments three times for each replicates, Classification was based on observation of color intensity and amount of precipitate.

Antimicrobial assay

The antimicrobial activities of methanol and ethanol extracts of *T. stans* gave different zones of inhibition on the organisms tested (Table 2). The methanolic and ethanolic extracts inhibited the growth of all most all the isolates of bacteria and fungi. The methanol and ethanol extract showed more potent against *E. coli*, *Xanthomonas axanopodis* pv. *malvacearum*, *Clavibacter michiganensis* sub sp. *michiganensis, Staphylococcus aureus, Klebsiella pneumoniae, Pseudomonas aeruginosa,Pseudomonas fluorescens* and moderate activity observed in *Xanthomonas oryzae* pv. *oryzae*. All the solvent extracts exhibited high activity on all species of *Aspergillus* and *Alternaria*. All the extracts did not showed any effect on species of *Fusarium* and *Nigrospora oryzae*.

Table 2: Zone of inhibition (in mm) of antimicrobial activity y disc diffusion method using different solvent extract of *Tecoma stans.*

Microorganisms	Samples			
Bacterial pathogens	**Methanol**	**Ethanol**	**water**	**chloramphenicol**
Klebsiella pneumonia	9±1	8±1	6±1	18±
Escherichia coli	13±2	12±2	3±1	20±
Staphylococcus aureus	10±1	9±1	1±1	18±2
Pseudomonas aeruginosa	9±1	7±1	4±1	18±
Clavibacter michiganensis sub sp. *michiganensis*	11±2	12±2	4±1	16±
Pseudomonas fluorescens	8±1	7±1	3±1	21±
Xanthomonas oryzae pv. *oryzae*	5±1	4±1	2±1	16±
Xanthomonas axanopodis pv. *malvacearum*	13±2	14±2	5±1	22±
Fungal pathogens	Carbendazim			
Aspergillus niger	14±2	12±2	4±1	21±
Aspergillus flavus	15±2	10±2	2±1	21±
Alternaria carthami	8±1	7±1	3±1	19±
Alternaria helianthi	5±1	5±1	2±1	21±
Cercospora carthami	7±1	5±1	3±1	21±
Nigrospora oryzae	5±1	4±1	2±1	20±

+: Presence; -: absence, repeated the each experiments three times for each replicates

DISCUSSION

In recent years, the search for phytochemicals possessing antimicrobial properties has been on the rise due to their potential use in the therapy of various chronic and infectious diseases. Due to risk of adverse effects encountered with the use of synthetic antibiotics, medicinal plants may offer an alternative source for antimicrobial agent with significant activity against pathogenic and infective microorganisms. In addition, a number of antibiotics have lost their effectiveness due to the development of resistant strains, mostly through the expression of resistance genes.

Results of our findings confirmed the use of *T. stans* as traditional medicine. We found strong antimicrobial and antioxidant activities specifically in the ethanolic and methanolic extracts of *T. stans*. High TPC values found in ethanolic and methanolic extracts (11.32 and 11.64 mg GAE/g extract) imply the role of phenolic compounds in contributing these activities.

Plant phenolic compounds have been found to possess potent antimicrobial[4].

The flavonoids from plant extracts have been found to possess antimicrobial properties in various studies[8, 9, 10, 11]. The presence of alkaloids has shown as antimicrobial activity[12]. Plant based steroids also possess as antimicrobial extracts[13].

The anthraquinones have also shown as antimicrobial properties[14]. Strong presence of tannins in all extracts may explain its potent bioactivities are known to possess potent antimicrobial activities[4]. The Saponins from plant extracts have already reported as potent antimicrobial [15]. The present investigation has shown that the ethanol and methanol extract of *T. stans* have active phytochemicals which are able to inhibit plant and animal pathogenic bacteria and fungi. The ethanol and methanol extract fractions showed significantly antimicrobial activity against all Gram-positive and Gram negative bacteria and different fungi tested. These findings provide scientific evidence to support traditional medicinal uses and indicate a promising potential for the development of an antimicrobial agent from *T. stans* plant. This medicinal plant by *in vitro* results appear as interesting and promising and may be effective as potential sources of novel antimicrobial drugs.

ACKNOWLEDGEMENT

The author feels to express sincere thanks to the Management, Principal and staff of PSGVP Mandal's college of Pharmacy, Shahada, The department of Pharmacology Karpagam University, Coimbatore, for their valuable support and co-operation during the research work.

REFERENCES

1. Fabricant, D.S. and Fansworth, N.R., The value of plants used in traditional medicine for drug discovery. *Environment Health Perspective,* 2001, 109, P. 69-75.
2. Prachayasittikul,S., Buraparuangsang, P.Worachartcheewan, A., Isarankura-Na-Ayudhya, C., Ruchirawat, S. and Prachayasittikul, V., Antimicrobial and antioxidant activity of bioreactive constituents from *Hydnophytum formicarum* Jack. *Molecules*, 2008, 13, P. 904-921.
3. Kumar, R.A., Sridevi, K., Kumar, N.V., Nanduri, S. and Rajagopal, S., Anticancer and immunostimulatory compounds from *Andrographis paniculata*. *J. Ethnopharm*, 2004, 92, P. 291-295.
4. Kaur, G.J. and Arora, D.S., Antibacterial and phytochemical screening of *Anethum graveolens, Foeniculum vulgare and Trachyspermum ammi*. *BMC Complement and Alternate Medicine*, 2009, 9, P. 30.
5. Halliwell, B., How to characterize an antioxidant: an update. *Biochemical Society Symposia*, 1995, 61, P. 85-91.
6. Senthilkumar, C.S., Sureshkumar, M. and Pandian, M.R., *In vitro* antibacterial activity of crude leaf extracts from *Tecoma stans* (L) Juss. ex Kunth, *Colues forskohlii* and *Pogostemon patchouli* against human pathogenic bacteria. *International Journal of PharmTech Research*, 2010, 2(1), P. 438-442.
7. Lai, H.Y., Yau, Y.Y. and Kim, K.H., *Blechnum orientale* Linn - a fern with potential as antioxidant, anticancer and antibacterial agent. *BMC Complementary and Alternative Medicine*, 2010, 10, P. 15.
8. Lin, Y., Shi, R., Wang, X. and Shen, H.M., Luteolin, a flavonoid with potential for cancer prevention and therapy**.** *Cure Cancer Drug Targets*, 2008, 8, P. 634-46.
9. Lopez-Lazaro, M., Distribution and biological activities of the flavonoid luteolin**.** *Mini Rev Med Chem,* 2009, 9, P. 31-59.
10. Yoshida, T., Konishi, M., Horinaka, M., Yasuda, T., Goda, A.E., Taniguchi, H., Yano, K., Wakada, M. and Sakai, T., Kaempferol sensitizes colon cancer cells to TRAIL-induced apoptosis**.** *Biochem and Biophys Res Comm*, 2008, 375, 129-133.
11. Amarlal, S., Mira, L., Nogueira, J.M., da Silva, A.P. and Florencio, M.H., Plant extracts with anti-inflammatory properties-a new approach for characterization of their bioactive compounds and establishment of structure-antioxidant activity relationships. *Bioorg. Med. Chem,* 2009, 17(5), P. 1876-1883.
12. Erdemoglu, N., Sozkanm S. and Tosun, F., Alkaloid profile and antimicrobial activity of *Lupinus angustifolius* L. alkaloid extract. *Phytochemistry Reviews,* 2007, 6(1), P. 197-201.
13. Shihabudeen, M.S., Priscilla, H.D. and Kavitha, T., Antimicrobial activity and phytochemical analysis of selected Indian folk medicinal plants. *International Journal of Pharma Sciences and Research*, 2010, 1(10), P. 430-434.
14. Comini, L.R., Nunez Montoya, S.C., Paez, P.L., Argüello, G.A., Albesa, I and Cabrera, J.L., Antibacterial activity of anthraquinone derivatives from *Heterophyllaea pustulata* (Rubiaceae). *J Photochem Photobiol B*, 2011, 102(2), P.108-114.
15. Mandal, P., Babu, S.S.P. and Mandal, N.C., Antimicrobial activity of saponins from *Acacia auriculiformis*. *Fitoterepia,* 2005, 76(5), P. 462-465.