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Alfa Universal
An International Journal of Chemistry
Vol 1 (2), 1-07 (2010)

Antimicrobial Activity and Phytochemical Screening on an Ornamental Foliage Plant, *Pothos aurea* (Linden ex Andre)

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(Received: March 16, 2010; Accepted: June 14, 2010)

Abstract: The *in vitro* antimicrobial activity of the petroleum ether, acetone and ethanolic extracts of the aerial roots of *Pothos aurea* (commonly known as money plant) that climbed on *Areca catechu* (betel nut palm) and *Lawsonia intermis* (Mehandhi plant) were investigated. The extracts exhibited antimicrobial activities with the zones of inhibition ranging from 0 to 15, 10 to 15 and 10 to 15 mm for petroleum ether, acetone and ethanol respectively. The extracts of the aerial roots from *Areca catechu* showed good antimicrobial activity. Phytochemical screening revealed the presence of alkaloids, flavonoids, tannins, glycosides and anthraquinone in the extracts of aerial roots intertwined over *Areca catechu* and in the extracts of aerial roots of *Pothos aurea* collected from *Lawsonia intermis*. The potential of the crude stem extracts of *Pothos aurea* to inhibit the growth of bacteria and fungi is a sign of its broad spectrum antimicrobial potential which may be used in the management of microbial infections.

Keywords: *Pothos aurea*, *Scindapsus aureus*, *Epipremnum aureum*, Silver vine, Devil's Ivy, Solomon Island's Ivy, Money plant, Antimicrobial activity.

INTRODUCTION

Plant kingdom represents an enormous reservoir of biologically active compounds and so far only a fraction of these plants have been assayed. Nearly 50% of the drugs used in medicine are of plant origin. It is important to use phytochemical methods to screen and analyze bioactive components, not only for the quality control of crude drugs, but also for the elucidation of their therapeutic mechanisms. The plants that possess therapeutic properties exert beneficial pharmacological effects and are generally designated as

“Medicinal plants”. Diseases that have been managed traditionally using medicinal plant include malaria, epilepsy, infantile, convulsion, diarrhea, and dysentery, fungal and bacterial infections [1].

Pothos aurea is a popular ornamental foliage plant used in hanging baskets and as a vine for totem poles: *Pothos*, *scindapsus aureus*, *Epipremnum aureum*, Silver vine, Devil’s Ivy, Solomon Island’s Ivy and money plant are some among the names by which *Pothos aurea* is mentioned. *Pothos aurea* (*Rhaphidophora aurea*) belongs to the family of Araceae. It is a popular house plant with numerous cultivars and efficient at removing indoor pollutants such as formaldehyde, xylene and benzene. *Pothos aurea* is a scrambler shrub and it can climb by means of aerial roots over the trees and plants which hook over the tree branches. Studies conducted in the past using *Pothos aurea* have shown that the plant exhibits a broad-spectrum antimicrobial activity against various pathogens [2],[3]. A study has been conducted to uncover the phytochemicals present in the aerial roots of *Pothos aurea* and to evaluate the antimicrobial effect of the extracts.

MATERIALS AND METHODS

Plant materials and preparation of extract

Aerial roots of *Pothos aurea* intertwined over *Areca Catechu* and *Lawsonia intermis* were chosen for the study. The aerial roots of *Pothos aurea* intertwined on *Areca Catechu* was collected from Palakkad District and the aerial roots of *Pothos aurea* intertwined over *Lawsonia Intermis* was collected from Coimbatore. Both the roots were identified by Dr J. Sudhakar, Botanical survey of India, Agricultural University, Coimbatore, India. The plant materials collected from *Areca Catechu* (betel nut palm) is denoted by I and which collected from *Lawsonia Intermis* is denoted by II. The aerial roots were shade-dried, pulverized and extracted with petroleum ether, acetone and ethanol for 6 hour separately. The extract was filtered and concentrated *in vacuo* to remove the solvents.

Phytochemical Screening Procedure

The phytochemical screening was done for Petroleum ether, Acetone and ethanol extracts of I and II through standard procedures as given in Table I

Table - 1

Test	Procedure	References
Test for alkaloids	Wagner’s Test Mayer’s Test:	[4], [5], [10]
Test for flavonoids	Lead acetate Test Conc.H ₂ SO ₄ Test	[4], [5], [6].
Test for Tannins	Braemer’s Test:	[4], [6]
Test for Steroids	Liebermann-Burchard Test	[4], [5], [6], [10]
Tests for Terpenoid		[4], [5], [6]
Test for Glycosides	Legal’s test:	[4], [6]

Test for Saponins	Foam test:	[4]
Test for Anthraquinone	Borntrager's test:	[5], [6], [10]
Test for fixed oils and fats	Oil stain test:	[5], [6], [10]
Test for Phenols	FeCl ₃ Test	[5], [6]
Test for Carbohydrates	Fehling's Test:	[4], [5], [10]
Test for Rotenone	Rotenone Test	[11]

Antimicrobial Testing

The potential of the crude stem extracts of *Pothos aurea* to inhibit the growth of bacteria and fungi were determined by suitable antimicrobial tests.

Test microorganisms

The test microorganisms used in the study (bacteria: *Escherichia coli* and *Staphylococcus aureus* and fungi: *Aspergillus's flavus* and *Candida albicans*) were obtained from Jebi Laboratory, Pollachi, India.

Composition of Culture Media

Preparation of Culture Media for Antibacterial Studies

Mueller-Hinton Medium.

Content	g/litre	Content	g/litre
Acid Hydrolysate of Casein	17.5gm	Starch	1.5gm
Beef Extract	2.0gm	Agar	17.0gm

The antibacterial activity of the crude extracts was determined in accordance with the Kirby-Bauer antimicrobial disk diffusion method. Mueller Hinton dehydrated media (38g) was dissolved in 1000 ml of purified filtered water and heated with frequent agitation. Media was sterilized at 121° C for 15 minutes and cooled to 45-50° C and dispensed into sterile petri dishes. The bacterial isolates were first grown in a nutrient broth for 18 hours before use and standardized to 0.5 Mc Farland standards (10^6 cfuml⁻¹). Two hundred micro liter of the standardized cell suspensions were spread on a Mueller Hinton medium. The inoculum was swabbed onto plate surface three times. Place appropriate susceptibility disk on the surface and press gently. Incubate at 35° C for 24 hours.

Preparation of cultural media for antifungal studies

SDA Medium (Sabouard Dextrose Agar)

Contents	g/litre
SDA	65 g
Distilled water	100 ml

Suspended 65.0 g of SDA in 100 ml distilled water and boiled to dissolve the medium completely. Sterilized by autoclaving at 15 lbs pressure and 121°C for 15 min.

Sterilization

The Petri dishes were sterilized by means of hot air oven at 160°C for 1 hour. Melted Mueller Hinton agar for bacteria and SDA for fungi, were poured into separate dishes to a uniform depth of 5 mm and refrigerated for solidification. The plates were then kept in an incubator at 37°C for 10 to 20 minutes to dry off the moisture that develop on the agar surface.

Antimicrobial testing

Disc diffusion method for Determination of Zone of Inhibition for Antimicrobial activity:

Paper discs of 4 mm diameter and glass Petri plates of 90 mm diameter were used throughout the experiment. Paper discs were sterilized in an autoclave and dried at 100°C in an oven. Then the discs were soaked with test chemicals at the rate of 50 µg (dry weight) per disc for antimicrobial analysis. One drop of bacterial suspension was taken in a sterile Petri dish and then approximately 20 ml of sterilized and melted Mueller Hinton agar (~45°C) was poured into the plate and then mixed thoroughly. Another one drop of microorganisms for antifungal activity was also taken in a sterile Petri dish and then sterilized and melted SDA was poured into the plate and then mixed thoroughly. The paper discs after soaking with test chemicals were placed at the center of the inoculated pour plate. A control plate was also maintained in case with alcohol. First, the plates were maintained at low temperature (4°C) for 4 hours. The plates were then incubated at (35±2)°C for growth of test organisms and were observed at 24 hour intervals for two days. The activity is expressed in terms of zone of inhibition in mm. Each experiment was repeated three times. Gentamycin was used as a positive control for antibacterial activity and Ketaconazole was taken as a positive control for antifungal activity and compare with test chemicals under identical conditions.

RESULTS AND DISCUSSION

Phytochemical screening revealed the presence of alkaloids, flavonoids, tannins, steroids and anthraquinone in petroleum ether extracts of I; alkaloid, tannin, glycosides anthraquinone and carbohydrates in acetone extracts of I and alkaloid, tannin, glycoside, anthraquinone and phenol in ethanol extracts of I.

Analysis of petroleum ether extracts of II revealed the presence of alkaloid, flavonoid, tannin, terpenoid and anthraquinone. Alkaloid, tannin, and glycosides were present in the acetone extracts of II and alkaloid, flavonoid, tannin, glycoside, saponin and anthraquinone were present in the ethanol extracts of II.

A comparison of petroleum ether extracts of I and II revealed that, steroids are present in I, but not in petroleum ether extracts of II. Terpenoid is present only in petroleum ether extract of II. Anthraquinones and carbohydrates are present in the acetone extract of I, while they are absent in acetone extract of II. Phenol is present in ethanol extract of I while it is absent in ethanol extract of II. Flavonoids and saponin are present in ethanol extract of II, whereas it is absent in ethanol extract of I. This may be attributed to the varying phytochemical constituents of the host tree.

Table –1(a)

Phytochemical screening of the Petroleum ether, Acetone and Ethanol extracts of *Pothos aurea* (I).

Phytochemicals	Petroleum Ether	Acetone	Ethanol
Alkaloids	+	+	+
Flavonoids	+	–	–
Tannin	+	+	+
Sterols	+	–	–
Terpenoid	–	–	–
Glycosides	–	+	+
Saponin	–	–	–
Anthraquinone	+	+	+
Fixed fat & oils	–	–	–
Phenols	–	–	+
Carbohydrates	–	+	–
Rotenone	–	–	–

Table –1(b)

Phytochemical screening of the Petroleum ether, Acetone and Ethanol extracts of *Pothos aurea* (II).

Phytochemicals	Petroleum Ether	Acetone	Ethanol
Alkaloids	+	+	+
Flavonoids	+	–	+
Tannin	+	+	+
Sterols	–	–	–
Terpenoid	+	–	–
Glycosides	–	+	+
Saponin	–	–	+
Anthraquinone	+	–	+
Fixed fat & oils	–	–	–
Phenols	–	–	–
Carbohydrates	–	–	–
Rotenone	–	–	–

Antibacterial Activity

Table – 1(c)

Extracts	Zone of inhibition (mm) against	
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>
Petroleum ether of I	10	15
Acetone of I	15	10
Ethanol of I	15	10
Petroleum ether of II	Resistant	Resistant
Acetone of II	10	15
Ethanol of II	10	10
*Gentamycin (STD)	25	35

The extracts obtained were screened for their antibacterial activity by Disc diffusion method using isopropyl alcohol as solvent. 50µg/ml solutions of each extracts were compared with standard drug Gentamycin. A clear zone of growth inhibition was noted around the disc due to diffusion of drug. The diameter of the inhibition zone denotes the relative susceptibility of the test microorganisms to a particular anti microbe [7].

It is well apparent from the measured values of zone of inhibition (Table-1(c)) that the extracts showed good sensitivity against *Escherichia coli* than that of *Staphylococcus aureus*. The acetone and ethanol extract of I showed high sensitivity against *Escherichia coli*. This may be due to the presence of active phytochemicals like alkaloid, tannin, glycoside, anthraquinone, carbohydrate and phenol. Petroleum ether extract of I, acetone and ethanol extract of II showed moderately good sensitivity against *Escherichia coli*. Petroleum ether extract of I and acetone extract of II showed good sensitivity against *staphylococcus aureus*, while acetone extract of II, ethanol extract of I and II showed moderately good sensitivity.

Antifungal Activity

Table-1(d)

Extracts	Zone of inhibition (mm) against	
	<i>Aspergillus flavus</i>	<i>Candida albicans</i>
Petroleum ether of I	10	10
Acetone of I	15	15
Ethanol of I	10	15

Petroleum ether of II	Resistant	Resistant
Acetone of II	15	15
Ethanol of II	10	10
Ketaconazole (STD)	20	20

The measured value of zone of inhibition (Table-1(d)) proved that the six extracts showed good sensitivity against *Candida albicans* than that for *Aspergillus flavus*. Acetone and ethanol extract of I, and acetone extract of II showed good sensitivity against *Candida albicans*. Petroleum ether extract of II was found to be resistant against both bacteria and fungi. This may be due to the absence of steroids and glycosides as active phytochemicals.

The measured value of zone of inhibition against both bacteria and fungi revealed that the acetone extracts of I and II to be more sensitive than petroleum ether extract of I and II. Acetone extract of I and II showed good sensitivity against fungi.

The antimicrobial tests for six extracts proved that they are most sensitive to the fungi, than that of bacteria by comparing the zone of inhibition for each extract against the particular microbe.

The analysis of the six extracts of I and II showed good and moderately good sensitivity respectively against the fungi and bacteria. This proves the potential of the crude stem extracts of *Pothos aurea* to inhibit the growth of bacteria and fungi is a sign of its broad spectrum antimicrobial potential which may be used in the management of microbial infections.

CONCLUSION

The phytochemical constituents in the aerial roots of *Pothos aurea* climbed over two different trees may vary. This may be attributed to the varying phytochemical constituents of the host tree. The results of this preliminary analysis gives evidence that the aerial roots of *Pothos aurea* can be regarded as promising sources for antimicrobial drugs. Further isolation of phytochemical constituents through column chromatography techniques are being carried out in our laboratory.

ACKNOWLEDGEMENT

The authors thank that the concerned authorities of Avinashilingam University for Women for providing facilities to carry out this work.

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