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Research Article

ANTI- BACTERIAL ACTIVITY OF LEAF OF *NYCTANTHES ARBORTRISTIS* LINN

DH. Geetha,* Indhiramuthu Jayashree and M. Rajeswari
PG and Research Department of Botany, Vellalar College for Women, (Autonomous), Erode- 638012, Tamil Nadu, India

*Corresponding Author: DH. Geetha; Email: geethadhandapani2010@gmail.com

Abstract: In the current study the ethanolic and aqueous extracts of leaf of *Nyctanthes arbortristis* were screened for phytochemicals and antibacterial activity using agar well diffusion assay. Streptomycin was used as positive control. The test microorganisms used in the present study were *Bacillus subtilis*, *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Proteus mirabilis*. Ethanol extracts showed higher degree of antibacterial activity than aqueous extracts.
Key words: *Nyctanthes arbortristis*, Agar well diffusion assay, Phytochemical, Streptomycin

INTRODUCTION

Nature has provided the storehouse of remedies to cure all ailments of mankind. There is no doubt that plants are a reservoir of potentially useful chemical compounds which serve as drugs, and provide newer leads for modern drug synthesis[1]. *Nyctanthes arbortristis* L. commonly known as Harsinger or Night jasmine, is a well-documented plant belonging to the family *Oleaceae*. It is a native of India, distributed wild in sub-Himalayan region and also found in Indian garden as ornamental plant. Juice of the leaves is used as digestives, antidote to reptile venoms, mild bitter tonic, laxative, enlargement of spleen, diaphoretic diuretic[2-4] and antimicrobial activities[5]. The whole plant is used for treatment of cancer, root for fever, sciatica, anorexia; bark as expectorant, leaf for control fever, diabetes and as cholagogue, diaphoretic and anthelmintic. Various extracts of the plant is used to treat arthritis [6-8], malaria, intestinal worms tonic, laxative, antitrypanosomal, anti-inflammatory and antioxidant activity [9-11]. The plants are very well known for their pharmacological properties since ancient age.

MATERIALS AND METHODS

Plant Collection and Extraction

The leaves of *Nyctanthes arbortristis* were collected from Thindal, Erode district and were authenticated and deposited at the PG and Research Department of Botany, Vellalar College for Women, Erode (Tamil Nadu), India. Fresh leaves were collected and air-dried at room temperature and then homogenized to obtain coarse powder. The powdered leaf was extracted[12] with the solvent ethanol by hot extraction using soxhlet apparatus, collected and stored in a vial for further analysis.

Phytochemical Screening

Ethanolic and aqueous extracts were subjected to qualitative phytochemical tests followed by the methods [13-14].

Antibacterial Assay

Agar Well Diffusion Method

The antibacterial activity of ethanolic and aqueous leaf extracts of *N.arbortristis* was evaluated by well diffusion method[15]. The inoculation of micro-organisms was prepared from bacterial culture. About 20ml of Muller Hinton agar medium was poured in a sterilized petridish and allowed to solidify. One drop of bacterial strain was spread over the medium by a rod. Wells of 5mm in diameter and about 2cm apart were punctured in the culture medium using sterile cork borers. Varying concentrations of the ethanol and aqueous leaf extracts (25, 50, 75 and 100µg/ml) was added to the wells separately and the plates were incubated at 37°C for 24h. Streptomycin (50µg/ml) the standard antibiotic was used as positive control. Antibacterial activities were assessed by measuring inhibition zone in diameters and the results were given in relative magnitude of inhibition RMI.

Microorganisms Tested

Bacterial Strains used in this study were purchased from Kovai Medical College Hospital (KMCH), Coimbatore. These are *Bacillus subtilis*, *Staphylococcus aureus* (Gram positive) and *Klebsiella pneumoniae*, *Proteus mirabilis* (Gram negative). Bacteria were cultured overnight at 37^0C in Muller and Hilton Broth (MHB) 72hours in Potato Dextrose Broth and used as inoculum.

RESULTS AND DISCUSSION

The results of phytochemical tests of the solvent extracts were tabulated in Table 1. The ethanol extract and aqueous extract showed the presence of tannins, flavonoids, alkaloids, saponins and triterpenoids. The antibacterial activity of the various solvent extract of *Nyctanthes arbortristis* shows significant variation as shown in Tables 2 and 3. The results clearly show that the plant extracts were specific in action against the growth of bacteria. The ethanol extract showed the maximum activity against *Staphylococcus aureus* and *Klebsiella pneumoniae* with RMI of 3.4cm^2 at 100µg/ml which was comparable to the standard drug (RMI= 3.6cm^2) at 50µg/ml concentration. On the other hand, the aqueous extract showed maximum effect

against *Proteus mirabilis* and *Klebsiella pneumoniae* with RMI of 3.2 and 3.4cm^2 respectively, at 100µg/ml.

Table 1: Phytochemical screening of various extracts of *Nyctanthes arbortristis*

S. No.	Phytoconstituents	Ethanolic extract	Water extract
1.	Tannins (Ferric Chloride test)	+	+
2.	Flavonoids (Alkaline reagent test)	-	+
3.	Alkaloids (Dragendorff's test)	+	+
4.	Saponins (Froth test)	+	+
5.	Triterpenoids (Libermann-Burchard's test)	-	+

Table 2: Antibacterial activity of ethanolic extract of leaf of *Nyctanthes arbortristis* (RMI-cm^2)

S. No.	Name of Bacteria	Concentration of leaf extract (µg/ml)														
		25			50			75			100			AB		
		A_1	A_2	RMI	A_1	A_2	RMI	A_1	A_2	RMI	A_1	A_2	RMI	A_1	A 2	RMI
1.	*Bacillus subtilis*	0.5	0.5	1.0	0.5	0.9	1.8	0.5	1.2	2.2	0.5	1.4	2.6	0.5	1.5	3.0
2.	*Proteus mirabilis*	0.5	0.7	1.4	0.5	0.8	1.6	0.5	1.2	2.4	0.5	1.6	2.8	0.5	1.6	3.2
3.	*Klebsiella pneumoniae*	0.5	1.1	2.2	0.5	1.3	2.6	0.5	1.5	3.0	0.5	1.7	3.4	0.5	1.8	3.6
4.	*Staphylococcus aureus*	0.5	1.2	2.4	0.5	1.4	2.8	0.5	1.6	3.2	0.5	1.7	3.4	0.5	1.8	3.6

A_1 = Area of well in cm^2 RMI = A_2 / A_1

A_2 = Area of zone of inhibition in cm^2 (including area of well) AB = Antibiotic (Streptomycin -50 µg/ml)

Table 3: Antibacterial activity of aqueous extract of leaf of *Nyctanthes arbortristis* (RMI-cm^2)

S. No.	Name of Bacteria	Concentration of leaf extract (µg/ml)														
		25			50			75			100			AB		
		A_1	A_2	RMI	A_1	A_2	RMI	A_1	A_2	RMI	A_1	A_2	RMI	A_1	A 2	RMI
1.	*Bacillus subtilis*	0.5	0.7	1.4	0.5	0.8	1.6	0.5	1.2	2.4	0.5	1.4	2.8	0.5	1.5	3.0
2.	*Proteus mirabilis*	0.5	0.9	1.8	0.5	1.1	2.2	0.5	1.4	2.8	0.5	1.6	3.2	0.5	1.7	3.4
3.	*Klebsiella pneumoniae*	0.5	0.6	1.2	0.5	0.8	1.6	0.5	1.0	2.0	0.5	1.5	3.0	0.5	1.6	3.2
4.	*Staphylococcus aureus*	0.5	0.7	1.4	0.5	0.9	1.8	0.5	1.2	2.4	0.5	1.4	2.8	0.5	1.5	3.0

A_1 = Area of well in cm^2 RMI = A_2 / A_1

A_2 = Area of zone of inhibition in cm^2 (including area of well) AB = Antibiotic (Streptomycin -50 µg/ml)

CONCLUSION

The present study exhibited the antibacterial effect of ethanol and aqueous extracts of *N.arbortristis* against some of the pathogenic bacteria. As a result it is` assured that these extracts can confidently inhibit the growth of these microorganisms there by preventing various diseases and provide safe, easy, effective and practical solutions to every day ailments leaving behind no toxins and creating a clean and pleasant atmosphere. The plant extracts may be used to discover bioactive natural products that may serve as basic source for the development of new antimicrobial compounds to overcome the problem of increasing resistance to known traditional antibiotics.

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