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Nyctanthes arbor-tristis Against Pathogenic Bacteria

Savita G. Aggarwal and Sanjay Goyal

ABSTRACT

Nature has provided a complete storehouse of knowledge of drug. Herbal drugs constitute a major part in all traditional systems of medicines. Since ancient times mankind has exploited nature for all kind of useful production and enjoyed the colors, flavors and fragrances of flowers, food etc. Rigveda, the book supplies curious information on this subject. Despite the importance of western medicines, towards the end of the 20th century there again began a revival of interest in traditional medicines not only in developing countries, but also in the developed countries. The resurgence of plant based medicine is mainly due to the increasing evidences of the health hazards associated with the indiscriminate use of the modern medicine such as antibiotic, steroids and other synthetic drug. The plants used in the traditional system of medicine of India and china are now receiving much scientific attention. With the continuous use of antibiotics, microorganisms have become resistant. So, it is necessary to evaluate, in a scientific base, the potential use of folk medicine for the treatment of infectious disease produced by common pathogens. Thus it was thought worthwhile to carry out the systematic chemical examination of *Nyctanthes arbor-tristis*. Studies had been conducted to evaluate the antimicrobial properties of leaves of *Nyctanthes arbor-tristis*. It belongs to the family verbenaceae. The plant material was collected from herbal local nursery, and was stored for further studies. The different solvent extracts were prepared on the basis of polarity. Phytochemical analytical tests were carried out for preliminary investigation. Antimicrobial activities were evaluated using pathogenic microbes. The different solvent extracts of test material showed marked antimicrobial activity against pathogenic microorganism. The results showed that the test plant material was susceptible to different microorganism.

Keywords: Health Hazards, Antibiotic, Phytochemical, Microorganism.

1. Introduction

Since ancient times mankind has exploited nature for all kind of useful production and enjoyed the colors, flavors and fragrances of flowers, food etc. The Rigveda^[1] which is the oldest book in the library of man supplies curious information on the subjects. Presently many fine chemicals are derived from plants and used as medicines, dyes, flavors, fragrances, insecticides etc. Despite the importance of western medicines, presently about 80% of the world population is using medicinal plants as their major source for medication in primary treatment^[2]. Towards the end of the 20th century there began a revival of interest in traditional medicines not only in developing countries, but also in the developed countries. The resurgence of plant based medicine is mainly due to the increasing evidences of the health hazards associated with the indiscriminate use of the modern medicine such as antibiotic, steroids and other synthetic drug^[3]. The wealth of uninvestigated material available is illustrated by the fact that WHO reported that natural product research elicited some 3500 new chemical structures of which more than 2600 were from higher plants. Antimicrobial potential of different medicinal plants was being studied all over the world^[4]. The plants used in the traditional system of medicine of India and china are now receiving much scientific attention. Plants produce certain chemicals which are normally toxic to bacteria^[5] and many plants have been investigated for the development of novel drug with therapeutic value^[6]. Undoubtedly the plant kingdom still holds many species of plants containing substance of medicinal value which have yet to be discovered large number of plant are constantly being screened for their possible pharmacological value. With the continuous use of antibiotics, microorganisms have become resistant^[7].

So, it is necessary to evaluate, in a scientific base, the potential use of folk medicine for the treatment of infectious disease produced by common pathogens. Natural microbial can be derived from barks, stems, leaves Flowers and fruits of plant, various animals' tissues or from microorganisms [8]. Phytochemicals are the secondary metabolites that have several sub groups possessing various bioactivities such as antioxidants, antimicrobial, antiviral, anticancer etc [9].

Nyctanthes arbor-tristis is small sacred ornamental tree known across the country for its fragment white flowers. Plant is commonly known as night jasmine or parijata. It belongs to the family Verbenaceae [10]. It is native of India, distributed widely in the Sub Himalayan regions and Southward to Godavari. It grows in Indo- Malayan region and distributed across Terai tracts as well as Burma and Ceylon.

Traditionally the powdered stem bark is given in rheumatic joint pain, in treatment of malaria [11] and also used as an expectorant. The bark is used for the treatment of snakebite and bronchitis. Juice of the leaves is used as digestives [12], antidote to reptile venoms, mild bitter tonic, laxative, diaphoretic and diuretic.

Thus it was thought worthwhile to carry out the systematic chemical examination of *Nyctanthes arbor-tristis*. Studies had been conducted to evaluate the antimicrobial properties of leaves of *Nyctanthes arbor-tristis*.

2. Material & Methods

2.1 Collection and Identification of Plant Material:

The leaves of *Nyctanthes arbor-tristis* was collected from Biotech nursery, Paundha Road, Nanda ki chowki, Dehradun. The plant material is identified and authenticated by Dr. R.P. Kala, Head, Department of Forestry, UCTBMS, and Dehradun. The collected material was dried in shade. The dried material was stored in air tight poly bag till further use.

2.2 Experimental Procedure:

(a) Solvent Extraction

The plant material (100 gm., O.D. basis) of *Nyctanthes arbor-tristis* was taken in soxhlet apparatus. The constituents were extracted on the basis of increasing polarity of solvent. Plant material was extracted with following solvents petroleum ether, chloroform, Ethanol, Benzene, hot water in soxhlet apparatus for 8-10 hours. After the completion the extracts were dried under pressure.

(b) Phytochemical Screening of Different Solvent Extract

Various solvent extracts were qualitatively tested for carbohydrates, fats and oils, alkaloids, glycosides, saponins, flavonoids and for steroids and terpenoids by usual tests.

(c) Test Organism Used:

In vitro antimicrobial activity of various solvent extracts of plant material was examined. The test organism *Escherichia coli* (EC), *Pseudomonas aeruginosa* (PA), *Salmonella typhi* (ST), *Klebsiella*

pneumoniae (KP) and *Aspergillus niger* (AN) were the different microbial strains obtained from Institute of Microbial Technology (IMTECH), Chandigarh, India. These were obtained from pure lab culture of Department of Microbiology, Uttaranchal College of Technology & Biomedical Sciences, Dehradun. The entire microorganism was maintained at 40 °C on nutrients agar slants.

(d) Determination of Antimicrobial Activity:

The antimicrobial activities of different solvent extracts were evaluated by agar well diffusion method using Muller-Hinton-Agar. The culture medium was inoculated with the microorganism separately suspended in nutrient broth. A total of 8mm diameter wells were punched into the agar and filled with plant extracts (1mg/ml) and solvent blanks (petroleum ether, chloroform, Ethanol, Benzene, hot water as the case may be). Standard antibiotic (Amoxicillin (A), concentration 1 mg/ml) was simultaneously used as positive control. The bacterial plates were then incubated at 37 °C for 18 hours. The antimicrobial activity was evaluated by measuring the diameter of zone of inhibition observed. The experiments were repeated thrice and the mean values are presented.

3. Results and Discussion

The results of phytochemical tests of various solvent extracts were tabulated in Table 1. Among the different extracts ethanol extract followed by hot water extract and chloroform extract show the presence of tannins, terpenoids, steroids, flavonoids, glycosides & saponins.

The antimicrobial activities of the various solvent extract of *Nyctanthes arbor-tristis* shows significant variation as shown in Table 2. The hot water and ethanolic extracts of the leaves showed maximum antimicrobial activity followed by chloroform extract. The maximum zone of inhibition was shown with the *Pseudomonas aeruginosa* (PA), *Salmonella typhi* (ST). The successful prediction of botanical compounds from plant material is largely dependent on the type of solvent used in the extraction procedure. Traditional healers use primarily water as the solvent but in present studies the plant extracts in organic solvent (ethanol) provided more consistent antimicrobial activity compared to those extracted in water. These observations can be rationalized in terms of the polarity of the compounds being extracted by each solvent and in addition to their intrinsic bioactivity, by their ability to dissolve or diffuse in the different media used in the assay. It is also quite possible that leaves of the test plant material that were less effective in this do not possess antibiotic properties, or the extracts may have contained antimicrobial constituents, just not in sufficient concentrations so as to be effective. It is also possible that the active chemical constituents were not soluble in ethanol or water. The drying process may have caused conformational changes to occur in some of the chemical constituents found in the test plants material.

Table 1: Phytochemical Screening of Various Solvent Extract of *Nyctanthes arbor-tristis*

Phytochemical test	Hot water extract	Ethanolic extract	Benzene extract	Petroleum Ether extract	Chloroform extract
Test for Carbohydrates					
Benedict's test	-	-	-	-	-
Molisch's test	-	-	-	-	-
Test for Alkaloids					
Hager's test	+	+	-	-	+
Dragendorff's test	+	+	-	-	+
Test for Glycoside					
Legal's Test	-	+	-	-	-
Test for Saponins					
Froth test	+	+	-	-	+
Test for Tannins					
Ferric Chloride test	+	+	-	-	+
Test for Flavonoids					
Alkaline reagent test	+	+	-	-	-
Test for Steroids					
Salkowski's	+	+	-	-	-
Test for Fixed Oil					
Stain Test	-	-	-	-	-

Table 2: Antimicrobial Activity of various Solvent Extracts of *Nyctanthes arbor-tristis*

S. No.	Plant Extracts	Zone of Inhibition (mm)				
		EC	PA	ST	KP	AN
1	Control	25	22	20	22	18
2	Hot water extract	15	20	18	18	15
3	Ethanol extract	13	15	12	14	12
4	Benzene extract	09	12	NZ	NZ	NZ
5	Petroleum Ether extract	NZ	NZ	NZ	NZ	NZ
6	Chloroform extract	12	10	12	NZ	NZ

*EC: *Escherichia coli*, PA: *Pseudomonas aeruginosa*, ST: *Salmonella typhi*,
 KP: *Klebsiella pneumoniae*, AN: *Aspergillus niger*

4. Conclusion

The different solvent extracts of test material showed marked antimicrobial activity against pathogenic microorganism. The results showed that the test plant material was susceptible to different microorganism. The variation in the susceptibility of the organism could be attributed to the intrinsic properties of the organism. *Nyctanthes arbor-tristis* possessed a broad spectrum of activity against a panel of bacteria responsible for some common microbial disease in human and in plants.

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