International Journal of Phytopharmacology

www.onlineijp.com

e- ISSN 0975 – 9328
Print ISSN 2229 – 7472

Research Article

ANTIMICROBIAL ACTIVITY OF NYCTANTHES ARBORTRISTIS LINN BARK

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ABSTRACT

Antimicrobial activity of ethanolic extract of leaves of *Nyctanthes arbor-tristis* Linn bark was examined against selective bacteria and moulds by using disc diffusion method. Ethanol extract was found to be active against seven out of ten tested bacterial strains viz *P. aeruginosa, E. coli, S. epidermidis,* S. *aureus, S. pyogenes, B. subtilis, and K. pneumoniae* with the zone of inhibition of 31mm, 30mm, 25mm, 24mm, 22mm, 20mm and 19mm respectively. On the other hand, the aqueous extract was effective against five out of ten tested bacterial strains viz*S. aureus, S. dysenteriae, K. pneumoniae, B. megaterium and S. typhi* with the zone of inhibition of 23mm, 19mm, 18mm, 16mm and 14mm respectively and The antifungal activities of the pet.ether and ethanol extracts compared favorably with that of standard antibiotic. Pet.ether extract was found to be active against *Candida albicans, Aspergillus niger, Penicillum notatum, Candida glaberata* with 26mm, 25mm, 24mm, and 19mm zone of inhibition respectively. Where in turn ethanol extract was found to be active against *Candida albicans, Penicillum notatum, Aspergillus niger* with 23mm, 23mm and 21mm zone of inhibition respectively.

Key words: *Nyctanthes arbor-tristis* Linn bark, Antibacterial *K. pneumoniae, Penicillumnotatum*.

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INTRODUCTION

Nyctanthes arbor-tristis Linn bark. belongs to the family Oleaceae of the order Jasmlnaceae. Its synonyms are Sephalika, Parijatham in Sanskrit, Harsingar in Hindi and Coral / Night Jasmine in English. It is having brilliant, highly fragrant flowers which are white and yellow, and do not expand till evening and which fall off about sunrise. Thus during the day the plant lose all its brightness, and hence is called 'the sad tree'. Nyctanthes means 'Night-flowering'. It is occurring wild in the Sub-Himalayan region, Madhya Pradesh and Southwards to Godavari. It is cultivated in gardens as ornamental plants It has traditionally been used as antibacterial, analgesic, antirheumatic and a remedy in obstinate sciatica. Its leaves and corolla are used for the medicinal purpose & are collected during autumn season (Dey PM and Harborne JB, 1858; Mohan JSS & Inamdar JA, 1983; George K & Geethamma S, 1984; Kshetrapal S & Tiagi YD, 1974; Kundu BC & De A, 1989; Kuriachen PM & Dave YS, 1989; Majumdar DN and Agrawal AK, 1986; Saxena RS *et al*, 1984)

In present work the antibacterial studies were performed using some of the common gram negative bacteria, gram positive bacteria and moulds.

Access this article online

Home page: http://onlineijp.com/

DOI: http://dx.doi.org/10.21276/ijp.2017.8.4.4

Quick Response code

Received:25.08.17 **Revised**:12.09.17 **Accepted**:22.09.17

MATERIALS AND METHODS

Test microorganisms

The test microorganisms used in this study was:

Bacterial strains

Gram positive: *Bacillus subtilis, Bacillus megaterium, Staphylococcus epidermidis, Staphylococcus aureus, Staphylococcus pyogenes.* Gram negative strains: *Escherichia coli, Shigella dysenteriae, Pseudomonas aeruginosa, Salmonella typhi, Klebsiella pneumoniae.*

Fungi

Trichophyton longifusis, Candida glaberata, Candida albicans, Penicillum notatum, Aspergillus niger, Aspergillus flavus, Microsporum canis were obtained from NCIM (national collection of industrial microorganisms, National chemical laboratory, pune). The bacterial isolates were first subcultured in a nutrient broth (Oxoid) and incubated at 37°C for 18 h while the fungal isolates were subcultured on a Sabouraud dextrose agar (SDA) (Oxoid) for 72 h at 25°C.

Screening for anti bacterial activity

The antibacterial activity of the crude extracts was determined in accordance with the agar-well diffusion method [12]. The bacterial isolates were first grown in a nutrient broth for 18 h before use and standardized to 0.5 McFarland standards (106 cfuml-1). Two hundred micro liter of the standardized cell suspensions were spread on a Mueller-Hinton agar (Oxoid). Wells were then bored into the agar using a sterile 6 mm diameter cork borer. Approximately 100 µl of the crude extract at 10 mgml-1 were introduced into the wells, allowed to stand at room temperature for about 2h and then incubated at 37°C. Controls were set up in parallel using the solvents that were used to reconstitute the extract. The plates were observed for zones of inhibition after 24 h. The effects were compared with those of ciprofloxacin a concentration of 1 mg/ml

Screening for anti fungal activity

The fungal isolates were allowed to grow on a Sabouraud dextrose agar (SDA) (Oxoid) at 25°C until they sporulated. The fungal spores were harvested after sporulation by pouring a mixture of sterile glycerol and distilled water to the surface of the plate and later scraped the spores with a sterile glass rod. The harvested fungal spores and bacterial isolates were standardized to an OD600nm of 0.1 before use. One hundred microliter of the standardized fungal spore suspension was evenly spread on the SDA (Oxoid) using a glass spreader. Wells were then bored into the agar media using a sterile 6 mm cork borer and the wells filled with the solution of the extract taking care not to allow spillage of the solution to the surface of the agar medium. The plates were allowed to stand on the laboratory bench for 1 h to allow for proper diffusion of the extract into the media. Plates were incubated at 25°C for 96 h and later observed for zones of inhibition. The effect of the extract on fungal isolates was compared with miconazole at a concentration of 1 mg/ml.

RESULT AND DISCUSSION

Screening for anti bacterial activity

All the four different extract of *Nyctanthes arbor-tristis*Linn showed varying degree of antibacterial activities against the test bacterial species (Table 1). The antibacterial activities of the ethanol and aqueous extracts compared favorably with that of standard antibiotic. Ethanol extract was found to be active against seven out of ten tested bacterial strains viz *P. aeruginosa, E. coli, S. epidermidis,* S. *aureus, S. pyogenes, B. subtilis, and K. pneumoniae* with the zone of inhibition of 31mm, 30mm, 25mm, 24mm, 22mm, 20mm and 19mm respectively. On the other hand, the aqueous extract was effective against five out of ten tested bacterial strains viz*S. aureus, S. dysenteriae, K. pneumoniae, B. megaterium and S. typhi* with the zone of inhibition of 23mm, 19mm, 18mm, 16mm and 14mm respectively.

Screening for anti fungal activity

All the four different extracts of *Nyctanthes arbor-tristis*Linn showed encouraging antifungal activities against the tested fungal species (Table 2). The antifungal activities of the pet.ether and ethanol extracts compared favorably with that of standard antibiotic. Pet.ether extract was found to be active against *Candida albicans, Aspergillus niger, Penicillum notatum, Candida glaberata* with 26mm, 25mm, 24mm, and 19mm zone of inhibition respectively. Where in turn ethanol extract was found to be active against *Candida albicans, Penicillum notatum, Aspergillus niger* with 23mm, 23mm and 21mm zone of inhibition respectively.

Today, there is a renewed interest in traditional medicine and an increasing demand for more drugs from plant sources. This revival of interest in plant-derived drugs is mainly due to the current widespread belief that green medicine is safe and more dependable than the costly synthetic drugs, many of which have adverse side effects (Anonymous; Suresh V *et al.,* 2011). The present study was to evaluate the different pet.ether, chloroform, ethanol and aqueous extracts of *Nyctanthes arbor tristis* Linn were evaluated for its antibacterial and antifungal activity against the different pathogens.

In screening for anti bacterial activity, among all tested Gram positive organisms *S. epidermidis, S. aureus*, in Gram negative organisms' *P. aeruginosa, E. coli* was more susceptible to the ethanol extract. Aqueous extract was active against S.*aureus* in Gram positive organisms and S. *dysenteriae, K. pneumoniae* in Gram negative organisms. (Fig -1, Fig-2)

In Screening for anti fungal activity, pet.ether extract was more susceptible to *Candida albicans, Aspergillus niger, Penicillum notatum, Candida glaberata* among the all tested fungal pathogens,

ethanolic extract was effective against *Candida albicans*, *Penicillum notatum*, *Aspergillus niger* among the all tested fungal pathogens. (Fig-3).

Investigations on the phytochemical screening of *Nyctanthes arbor-tristis* Linn bark extracts revealed the presence of alkaloids, Steroids, glycosides, flavanoids, saponins, phenolic compounds and tannins. These compounds are known to be biologically active and therefore aid the antimicrobial activities of the plant.

Tannins are known for their astringent property and anti microbial activity. One of the most common biological properties of alkaloids is its toxicity against cells of foreign organisms. Steroidal extracts from some medicinal plants which exhibited antibacterial activities on some bacterial isolates. Further study is under process to find out the exact phytoconstituents responsible for the anti microbial activity, to determine the mechanism of action of action.

As an antimicrobial agent, If other related further studies and clinical trials are carried out, it will definitely open up a new vistas in modern medicine.

Table 1. Anti bacterial activity of different extracts of *Nyctanthes arbor tristis* Linn bark

Tested bacterial strains	Diameter of zone of inhibition (mm)				
	Standard drug	Pet. Ether extract	Chloroform extract	Ethanol extract	Aqueous extract
Bacillus subtilis,	22	4	9	20	-
Bacillus megaterium	17	-	-	-	14
Staphylococcus epidermidis	29	7	-	25	-
Staphylococcus aureus	28	5	12	24	23
Staphylococcus pyogenes	-	-	-	22	-
Escherichia coli	31	10	7	30	-
Shigella dysenteriae	25	9	0	0	19
Pseudomonas aeruginosa	32	6	9	31	-
Salmonella typhi	-	-	-	-	16
Klebsiella pneumoniae	21	3	8	19	18

Table 2. Anti fungal activity of different extracts of *Nyctanthes arbor tristis* Linn bark

Tested fungal strains	Diameter of zone of inhibition (mm)				
	Standard drug	Pet.Ether extract	Chloroform extract	Ethanol extract	Aqueous extract
Trichophytonlongifusis	24	4	-	4	-
Candida glaberata	21	19	7	3	-
Candida albicans	28	26	8	23	6
Penicillumnotatum	25	24	10	23	5
Aspergillusniger	26	25	9	21	8
Aspergillusflavus	23	5	-	-	-
Microsporumcanis	19	3	-	2	-

Fig 1. Anti bacterial activity of different extracts of *Nyctanthes arbor tristis* Linn bark on Gram positive bacterial strains

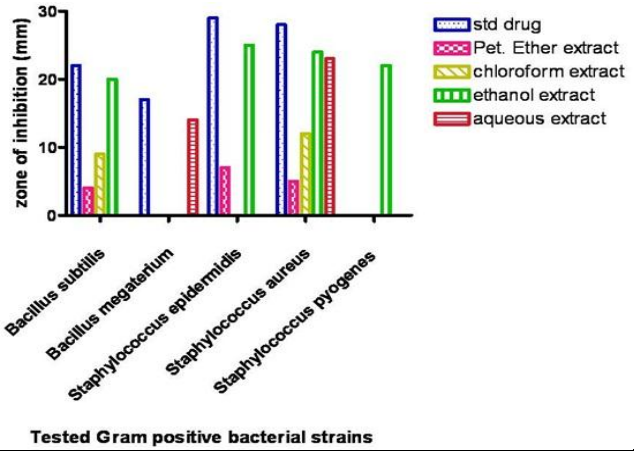


Fig 2. Anti bacterial activity of different extracts of *Nyctanthes arbor tristis* Linn bark on Gram negative bacterial strains

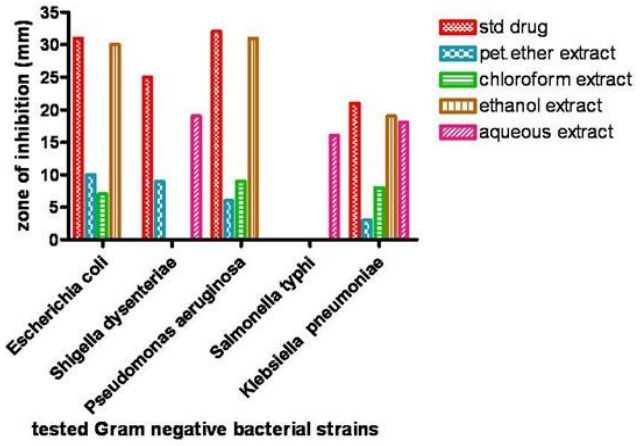
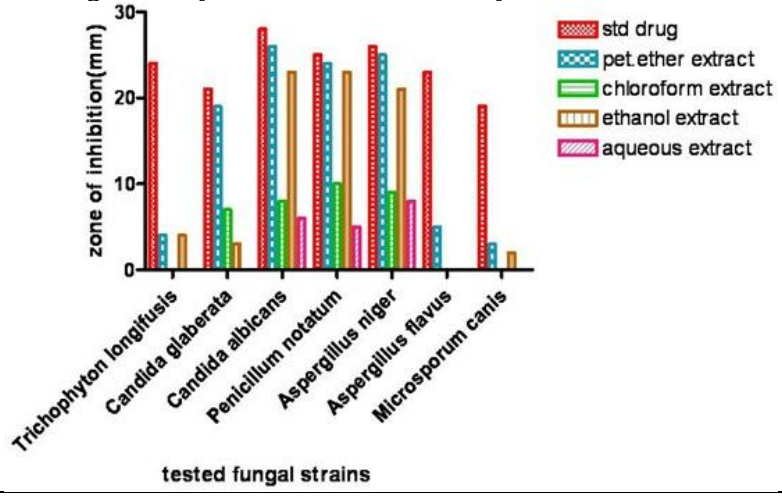


Fig 3. Anti fungal activity of different extracts of *Nyctanthes arbor tristis* Linn bark



CONCLUSION

Present study reveals that leaves of *Nyctanthes arbor-tristis* Linn has significant antimicrobial activity on selected strain. This study supports to identify exact mechanism and key phytochemical responsible to antimicrobial activity of *Nyctanthes arbor-tristis* Linn. it also helps to provide a way to identification potency of various parts of *Nyctanthes arbor-tristis* Linn. It also justifies the traditional usage of this plant as health remedy. The future prospect for effective utilization of this plant for the development of novel antibiotics will however depend on identification of the various chemical components of the phytoconstituents, purification of the components and the determination of their toxicity level with a view to establishing the biosafety of the plant as source of drug for human consumption.

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Cite this article:
Suresh V and Senthil Kumar N. Antimicrobial Activity of *Nyctanthes arbortristis Linn* Bark. *International Journal of Phytopharmacology*, 2017; 8(4):166-169. DOI: http://dx.doi.org/10.21276/ijp.2017.8.4.3