



## Research Article

### ANTIMICROBIAL STUDIES ON *NERIUM OLEANDER* LINN. LEAVES (WHITE KANER LEAVES)

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#### ABSTRACT

*Nerium oleander* is commonly known as Kaner (Hindi) found throughout in India and has been used in the treatment of cancer, leprosy, cardiac diseases and skin diseases. Various parts of plant roots, stems and leaves are used. The present study is therefore undertaken to analyse its antibacterial and antifungal activity in the chloroform and methanolic extract of leaves which were prepared by cold maceration method. These extracts were screened for antibacterial and antifungal activity by well diffusion method. Various concentrations (5µg, 10µg, 20µg, 40µg/ml) of extracts were used for antibacterial and antifungal studies. Out of four bacterial cultures used *E. coli* and *B. subtilis* showed better zone of inhibition in chloroform extract than methanolic extract. *Aspergillus brasiliensis* and *Candida albicans* are used for antifungal studies. Both the extracts show antifungal activity against these cultures.

**Keywords:** Nerium, Kaner, well diffusion, antibacterial, antifungal, zone of inhibition

#### INTRODUCTION

##### General Description

*Nerium oleander* is an evergreen shrub or small tree in the dogbane family Apocyanaceae. It is known as oleander from its superficial resemblance to the unrelated plant *Olive olea* but has many other names like *Nerium indicum* mill. and *Nerium odorum* soland. The white and red flowered variety is equated with *Nerium indicum*<sup>1</sup>.

##### Distribution

*N. oleander* is distributed in Mediterranean region and subtropical Asia, is indigenous to India–Pakistan subcontinent. Distributed in the Himalayas from Nepal westwards to Kashmir up to 1950m, extending to Baluchistan, Afghanistan and found throughout India in gardens. The white and red flowered variety is equated with *Nerium indicum*.

##### Classical names

Ayurvedic- Karavira, Viraka, Ashvamarka, Hayamaara, Gauripushpa, Siddhapushpa (white flower variety), Raktapushpa, Raktaprasava, Ravipriya (Red flowered variety)  
Unani- Kaner, Diflaa, Samm-ul-maar, Khar-Zaharah  
Sidha- Alari, Arabivayr  
English- Indian Oleander

##### Parts used

Leaves, roots, root bark

##### Classical uses

Charaka prescribed the leaves of white flowered variety externally in chronic and obstinate skin diseases of serious nature including leprosy. Sushruta used karavira in medicinal paste for application in alopecia. Root powdered with water was applied to alleviate venereal diseases. The powder of leaves was used as a snuff for treating epilepsy. All parts of plant especially roots were known to be highly poisonous when taken internally.

Tincture of flowers exhibited cardiotoxic, root CNS-active and spasmolytic activity. Externally, root exhibited healing properties for hemorrhoids and ulcers. Oil of rootbark gave good results in leprosy.

In Homoeopathy, tincture of *Nerium oleander* (red laurel) leaves is used in diseases of nervous system, hemiplegia and paralytic conditions under strict medical supervision.

Roots give plumericin, alpha-amyrin, beta-sitosterol, kaempferol, cardioactive glycosides named Odorosides A-H obtained from the root bark. Leaves contain the cardiac glycosides kaneroside, neriumoside, digitoxigenin, alpha –L-olendroside -5α-adynerin and other glycosides. Odorosides are cardioactive glycosides. Gentiobiosyl – oleandrin, Odoroside A and Oleandrin were the main glycosides identified<sup>2</sup>.

#### MATERIALS AND METHODS

##### Collection of plant material

*Nerium oleander* leaves of white variety were collected from the local Nangal area in Punjab. Sample of leaves was authenticated from the Director, NISCAIR New Delhi. The leaves were dried

under shade, coarsely grounded and kept at a lab temperature until used.

### Preparation for extract

The powdered leaf material was extracted by cold maceration method in Chloroform and Methanol. Extracted *Nerium oleander* leaves (LC AND MLO) extract was prepared in 5µg/ml, 10µg/ml, 20µg/ml and 40µg/ml conc. Then extracts were kept in pre-sterilized micro-centrifuge vials and stored at 4°C for study of antibacterial and antifungal activity. MLO— (Methanolic leaf extract) and LC (leaf chloroform extract)<sup>3</sup>.

### Selection of bacterial and fungal species

*Bacillus subtilis*, *Escherchia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* were taken for antibacterial studies and *Candida albicans* and *Aspergillus brasiliensis* were taken for antifungal studies.<sup>4,5</sup>

**Antibacterial and antifungal screening** of extracts was done by well diffusion method. Required glass ware was washed and dried in a hot air oven. The sterilized nutrient agar medium was

transferred to the Petri dishes, was allowed to solidify at room temperature. The selected test organism (Bacteria and fungi) was spread over the solidified agar with the help of a swab stick. Sterile borer was used to make wells of 8mm diameter. The dilutions of methanolic and chloroform leaves extract were prepared and put in the wells with the help of a sterile syringe needle. The Petri plates were placed in a refrigerator for 5min to allow diffusion. Later the Petri plates were incubated in inverted position at 37°C for 24 hours in the incubator. After 24hours the zone of inhibition was observed and diameter in mm was measured and recorded.<sup>6</sup>

### RESULTS

Positive results were shown by the presence of clear zones of inhibition around the active extracts. The zone of inhibition was measured and recorded using zone reader. The results obtained were compared with that of zone of inhibition produced by standard antibiotic. Out of different concentrations used 10µg/ml and 20µg/ml give better results.<sup>7</sup> The results were tabulated in Table 1.

**Table 1: Zone of inhibition in Bacterial culture & Fungal culture**

| Sl. No | Component /Extract       | Conc. (µg/ml) | Zone of inhibition for Bacteria |               |                  |                      | Zone of inhibition for Fungi |                        |
|--------|--------------------------|---------------|---------------------------------|---------------|------------------|----------------------|------------------------------|------------------------|
|        |                          |               | <i>B.Subtilis</i>               | <i>E.coli</i> | <i>S. aureus</i> | <i>P. aeruginosa</i> | <i>C. albicans</i>           | <i>A. brasiliensis</i> |
| 1      | LC (Chloroform extract)  | 5             | 11mm                            | 12mm          | -                | -                    | 19mm                         | 18.5mm                 |
|        |                          | 10            | 13mm                            | 15mm          | -                | --                   | 21 mm                        | 19 mm                  |
|        |                          | 20            | 15mm                            | 13mm          | -                | --                   | 22.5 mm                      | 19.3 mm                |
|        |                          | 40            | 15mm                            | 12mm          | --               | --                   | 22 mm                        | 21 mm                  |
| 2      | MLO (Methanolic extract) | 5             | -                               | -             | --               | --                   | --                           | -                      |
|        |                          | 10            | 11mm                            | 12mm          | --               | --                   | 13mm                         | 21mm                   |
|        |                          | 20            | 12mm                            | 12mm-         | -                | --                   | 15mm                         | 22mm                   |
|        |                          | 40            | 12mm                            | 12mm          | --               | --                   | 13mm                         | 21mm                   |

### DISCUSSION

Since the plant contain different phytoconstituents, antibacterial and antifungal studies were carried out to find activity against selected bacterial and antifungal species. Chloroform extract show better zone of inhibition (15 mm) out of two extracts. Out of four species of bacteria analysed *E. coli* and *B.subtilis* showed better zone of inhibition(15mm,15mm respectively). In antifungal studies both extracts show same zone of inhibition 22.5 and 22 respectively. *A.brasiliensis* showed better zone of inhibition as compared to *C.albicans*. This show that leaves of *Nerium oleander* has certain phytoconstituents which are responsible for antibacterial and antifungal activity<sup>8</sup>.

Since the growth of bacteria *Bacillus subtilis*, *E. coli*, *Pseudomonas aeruginosa* and *S.aureus* and fungi *Candida albicans* and *Aspergillus brasiliensis* controlled by *Nerium oleander*, it shows that they could inhibit the activity of bacteria and fungi which causes various diseases<sup>9</sup>.

### CONCLUSION

Since the plant has certain active constituents and is effective in controlling the growth of bacteria *Bacillus subtilis* and *E. coli*, and fungi *Candida albicans* and *Aspergillus brasiliensis*. Therefore, further studies can be carried out to isolate the active principle of plant, so a drug formulation could be made against these pathogenic bacteria.

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