



RESEARCH ARTICLE

Antibacterial Activity of *Gardenia gummifera* Linn. Extracts

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ABSTRACT

Gardenia gummifera Linn. belongs to family Rubiaceae. It is commonly known as Dikamali. Preliminary phytochemical screening of *G. gummifera* extract showed the presence of various phytoconstituents like glycosides, alkaloids, steroids, flavonoids, phenols, tannins and terpenoids. *Gardenia* species, such as *G. aqualla* have been reported to possess significant anti-microbial activity. Present study aimed at evaluating antibacterial activity of ethanolic and chloroform extracts of *G.gummifera* using agar well diffusion method. The activity spectrum was tested against different types of Gram positive (*B. subtilis* and *S. aureus*) and Gram negative (*E. coli* and *P. aeruginosa*) bacterial strains. The activity of both the extracts was compared using Amoxycillin as standard. Chloroform extract showed significant activity against all pathogens when compared with ethanolic extract of *G. gummifera*. The results observed in the present study indicate that the chloroform extract of the plant can be explored further to develop potential antimicrobial agents.

KEYWORDS

Gardenia Gummifera, Agar Diffusion Method, Zone of Inhibition

INTRODUCTION

Plants synthesize substances that provide protection against attack by insects, herbivores and microorganisms. It is due to the presence of substances such as tannins, phenolic compounds, alkaloids & so on.¹ Medicinal plants represent a rich source of antimicrobial agents. The different parts of plants used for medicinal purpose include root, stem, flower, fruit, seeds and twigs exudates. Large number of plants have been tested for their antibacterial activity and most of them are found effective against pathogenic micro-organisms.² With development in science and technology, tremendous progress has been made in the field of medicines.

Antibiotics are the most important therapeutic discovery of the 20th century.^{3,4} Resistance to the antibiotics have been increased in recent years.^{5,6}

Medicinal plants have been used in different traditional systems of medicine to treat numerous human diseases, hence researchers are now paying more attention in the isolation of biologically active components from the plants for the development of novel drug.^{3,7}

Gardenia gummifera Linn. belongs to family Rubiaceae, which is commonly known as Dikamali.

Traditionally, this medicinal plant, reported to have a number of therapeutic uses and can be used for its anthelmintic, antispasmodic, carminative, diaphoretic, expectorant, antiepileptic, peripheral and central analgesic, anti-oxidant and antihyperlipidemic activities.⁸

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MATERIAL AND METHODS

Collection and Authentication of Plant

The twigs of *G. gummifera* were procured from the Keshavshrushti, Bhayandar, Maharashtra. Plant was authenticated at Department of Botany, Guru Nanak Khalsa College, Matunga, Mumbai and voucher specimen (kw030815) was deposited for further reference.

Preparation of Plant Extracts

The twigs were shade dried and made into fine powder. Powdered material was subjected soxhlet extraction for 48 hrs using chloroform and ethanol as solvents. The extracts were concentrated under vacuum in rotary evaporator and dried completely. Both the extracts were stored in sterile glass bottle in refrigerator until use.

Culture Medium

Muller-Hinton agar medium was used to study the antibacterial activity of the chloroform and ethanolic extract of *G. gummifera*.

Microbial Culture

Four bacterial strains were used throughout investigation, two gram positive bacteria (*S. aureus* and *B. Subtilis*) and two gram negative bacteria (*E. Coli* and *P. Aeruginosa*). All the bacterial cultures were obtained from Microbiology Laboratory, Bharati Vidyapeeth's College of Pharmacy. These cultures were subcultured in nutrient agar media and used for the bioassay. The slants were prepared from the pure cultures and use further.

Inoculums Preparation

Cultures of above mentioned bacterial strains were removed from nutrient agar slant and transferred to fresh Muller-Hinton broth and incubated at 37 °C for 24hrs.

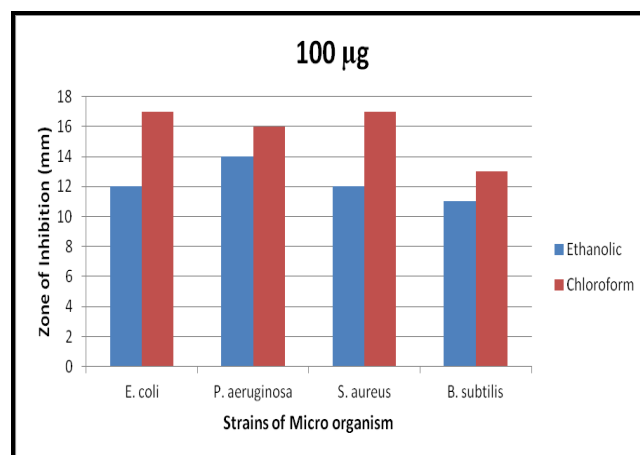
Antibacterial Assay

Agar well diffusion method was used to evaluate antibacterial activity. Muller-Hinton agar medium was prepared and sterilised in an autoclave at 121°C for 20 minutes at 15lbs psi then it was transferred into previously sterilised

glass petriplates. After solidification of agar, 0.1 ml of 24hrs old inoculums suspensions of *S.aureus*, *B. Subtilis*, *E. coli* and *P.aeruginosa* were poured on Muller- Hinton agar medium and spread throughout the plate by spread plate technique. Wells (8mm diameter) were punched in each of the plate using sterile borer. Two working concentrations, 1mg/ml and 2mg/ml of ethanolic and chloroform extracts were prepared and 100µl of all the solutions were added with sterile micro pipette into the respective wells and allowed to diffuse at room temperature for 2 hrs. Control experiment comprising inoculums without plant extract were also set up. The plates were incubated at 37°C for 24 hrs. The diameter of zone of inhibition (mm) was measured. Amoxycillin, 500µg/ml was used as reference standard.

RESULTS AND DISCUSSION

The antibacterial potential of both the extracts were evaluated on the basis of their zone of inhibition against four pathogens and the results were compared with the activity of the standard, Amoxycillin (500µg/ml). The results revealed that both the extracts have potent antibacterial activity against all the microorganisms studied. Among chloroform and ethanolic extract, chloroform extract showed significance activity than the ethanolic extract. In chloroform extract, maximum inhibitory zone was obtained against *E. coli* with diameter of 20 mm and least activity was seen against *B. subtilis*.

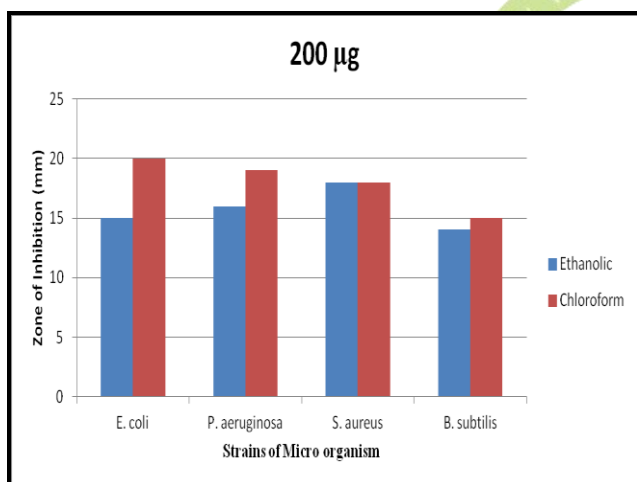


Graph 1: Comparative View of Inhibition Zone Made by Ethanolic and Chloroform Extract on Different Bacterial Strains at Conc. of 100µg

Table 1: Antibacterial Activity (Zone of Inhibition, mm) of Ethanolic and Chloroform Extracts of *G. gummifera* Against Clinical Pathogens

Bacterial strain	Zone of inhibition(mm)					
	Ethanolic		Standard	Chloroform		Standard
	100µg	200 µg	50 µg	100 µg	200 µg	50µg
<i>E. coli</i>	12	15	14	17	20	14
<i>P. aeruginosa</i>	14	16	14	16	19	14
<i>B. subtilis</i>	11	14	15	13	15	15
<i>S. aureus</i>	12	18	13	17	18	13
8mm diameter well						

Similarly, ethanolic extract showed maximum zone of inhibition with diameter of 18mm against *S. Aureus* and lowest in *B. subtilis*.



Graph 2: Comparative View of Inhibition Zone Made by Ethanolic and Chloroform Extracts on Different Bacterial Strains at Conc. of 200µg

In the present study, ethanolic and chloroform extracts of *Gardenia gummifera* were screened for antibacterial activity and compared with standard drug, Amoxycillin. It is evident from the data presented in Table 1 that both the extracts possess antibacterial activity. The agar well diffusion method result showed the zone of inhibition of 12mm, 14mm, 11mm, 12 mm for 100µg and 15mm,16mm,14mm,18mm for 200µg for ethanolic extract and 17 mm, 16 mm, 13 mm, 17 mm for 100 µg and 20 mm, 19 mm, 15 mm

and 18 mm for 200 µg of chloroform extract for the test sample against *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus substilis* and *Staphyllus aureus* respectively when compared with the standard drug Amoxycillin showing 14mm, 14 mm, 15 mm and 13 mm zone of inhibition respectively.

On the basis of results obtained from present study, it can be conclude that chloroform extract of *G. gummifera* possess significant antibacterial activity. Zone of inhibition is almost equal to the standard, which shows that the test organisms are sensitive to the plant extract.

Antibacterial Activity of Ethanolic and Chloroform Extract of *Gardenia gummifera*.

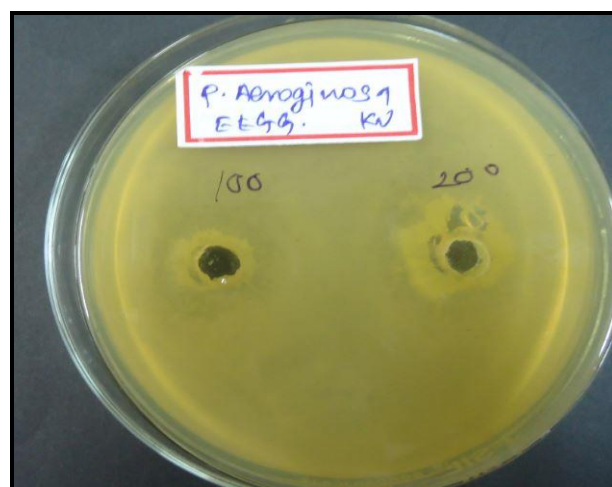


Figure 1: *P. aeruginosa* Ethanolic Extract 100µg and 200 µg

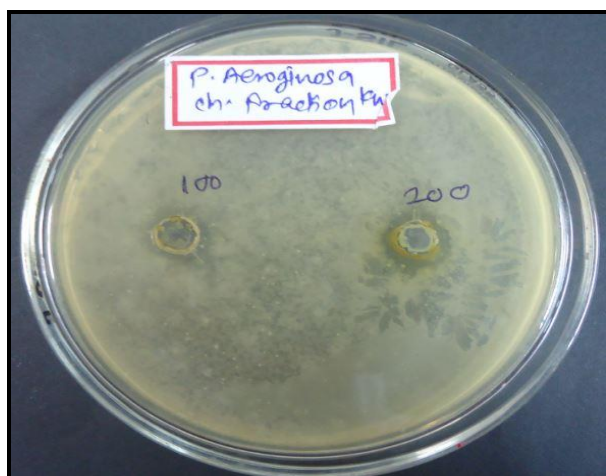


Figure 2: *P. aeruginosa* Chloroform Extract 100µg and 200 µg



Figure 3: *S. aureus* Ethanollic Extract 100µg and 200 µg



Figure 4: *B. subtilis* Chloroform Extract 100µg and 200 µg

CONCLUSION

The present investigation showed that, *G. gummifera* contain potential antibacterial components that can be used in the development of novel anti-microbial agents for the treatment of various infections. The ethanol and chloroform extracts of *G. gummifera* possess significant inhibitory effect against tested pathogens. Thus present study can be extended further by carrying out the isolation and purification of the antibacterial components from the active crude extract and their characterisation using analytical techniques such as – IR, NMR and MS.

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