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Anti-bacterial and phytochemical studies on methanolic extracts of *Begonia floccifera* Bedd. flower

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ABSTRACT

Objective: To investigate the antibacterial activity and phytochemical properties of *Begonia floccifera* Bedd. (*B. floccifera*) methanolic flower extracts against the selected pathogens. **Methods:** 20 g of fresh flowers were soaked in 100 mL methanol for 48 h at room temperature. After 48 h, the extracts were filtered using Whatman No. 41 filter paper. The filtrates were collected, made up to known volume and stored in refrigerator at 4 ℃. The methanolic extracts were used for phytochemical and antibacterial studies. The preliminary phytochemical screening was performed according to the modified Harborne method. The methanolic flower extracts of *B. floccifera* Bedd. was tested against *Escherichia coli, Klebsiella pneumonia, Pseudomonas aeruginosa, Staphylococcus aureus, Bacillus cereus, Salmonella typhi, Serratia marcescens, Proteus mirabilis, Enterococcus faecalis* and *Streptococcus pyogenes* by the disc diffusion method. **Results:** The results of the phytochemical screening revealed that phenol, tannins, xanthoproteins, steroids, tannins, steroids, phytosterols, triterpenoids, sapogenins, coumarins and carbohydrates presence in the methanolic extracts of *B. floccifera*. The antibacterial activity has been observed in the methanolic extracts of *B. floccifera* against the tested bacteria with varied activity. The maximum zone of inhibition was 28 mm for Bacillus cereus, 25 mm for Staphylococcus aureus, 15 mm for Escherichia coli, 13 mm for Proteus mirabilis, 7 mm for *Klebsiella pneumonia*. **Conclusions:** The antimicrobial activity of *B. floccifera* methanolic flower extracts are comparable and their potential as alternative in the treatment of infectious by these microorganisms.

1. Introduction

Tropical forests are the major reservoirs of plant diversity and inhabit a large number of medicinal, aromatic and wild edible plants [1-2]. The edible plants collected from wild are not only dietary staples but also provide more nutritionally valuable supplements as they are rich in vital nutrients [3]. Medicinal plants are an important source of inexpensive and practical drugs for people throughout the world. Medicinal plants are plants which contain thousands of substances that could be used for therapeutic purposes or which are precursors for the synthesis of useful drugs. The wealth of the medicinal plants in India especially South India has led us to an escalating curiosity in the exploration of ethnomedicinal plants as potential source of new antimicrobial agents. The herbal products today symbolise safety in contrast to the synthetics that are regarded as unsafe to human and environment. Although herbs had been priced for their medicinal, flavouring and aromatic qualities for centuries, the synthetic products of the modern age surpassed their importance, for a while. However, the blind dependence on synthetics is over and people are returning to the naturals with hope of safety and security [4-5]. Recent studies are involved in the identification and isolation of new therapeutic compounds of medicinal importance from higher plants for specific diseases [6-10]. In recent years, pharmaceutical companies have spent considerable time and money in developing therapeutics based upon natural products extracted from plants [11-12]. *Begonia floccifera* Bedd. (*B. floccifera*) belongs to the family Begoniaceae is endemic to Western Ghats and being used as a wild edible by the Kani tribe of Kanyakumari district [13]. Traditionally leaves of this plant are used to cure venereal diseases and

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give cooling effects to the body among the tribal inhabitants of Tirunelveli Hills of Western Ghats [14]. Having the potent edible and medicinal uses, this plant has not yet been subjected to phytochemistry and bio-efficacy studies. Thus the present study was aimed to investigate the preliminary phytochemical and antibacterial studies on this endemic species *B. floccifera* Bedd. (Begoniaceae), which is confined to Western Ghats, especially the hills of Travancore and Tirunelveli.

2. Materials and methods

Fresh flowers of *B. floccifera* Bedd were plucked from the wild habitat (Kalial range, Western Ghats of Kanyakumari, Tamil Nadu, India). 20 g of fresh flowers were soaked in 100 mL methanol for 48 h at room temperature. After 48 h, the extracts were filtered using Whatman No. 41 filter paper. The filtrates were collected, made up to known volume and stored in refrigerator at 4 ℃. The methanolic extracts were used for phytochemical and antibacterial studies. The preliminary phytochemical screening was performed according to the modified Harborne [15-19] method. The methanolic extracts of *B. floccifera* were tested against a panel of microorganisms including *Escherichia coli, Klebsiella pneumonia, Pseudomonas aeruginosa, Staphylococcus aureus, Bacillus cereus, Salmonella typhi, Serratia marcescens, Proteus mirabilis, Enterococcus faecalis* and *Streptococcus pyogenes* obtained from ATCC, Chandigarh. The antimicrobial activity was tested through disc diffusion method [20]. Nutrient agar was used as the standard test medium for the present study. Fresh cultures were prepared and used to inoculate 50 ml of Muller- Hinton broth that was incubated at 35 ℃ for 18 h. Overnight broth cultures were prepared, adjusted in peptone-physiological salt solution (1 g peptone and 8.5 g/L NaCl) to yield approximately 106 bacteria/mL. The agar plates were prepared in 90 mm Petri dishes with 22 mL of agar medium giving a final depth of 3 mm. Methanolic extracts (125 μg/mL) loaded discs were placed on the inoculated agar surfaces. Methanol (75%) was used as negative control. Amikacin (30 μg) was used as positive controls. All plates were aseptically incubated at 37 ℃ for 18-24 h. The antimicrobial activity was estimated by measuring the radius of the inhibition zone (mm). Each test was performed in triplicate and the results were shown as means.

3. Results

Methanolic flower extracts of *B. floccifera* were examined for the phyto-constituents presence and antibacterial activity against the human pathogens and the results are given in the table - 1 and 2. The results of the phytochemical screening revealed that phenol, tannins, xanthoproteins, steroids, phytosterols, triterpenoids, sapogenins, coumarins and carbohydrates presence in the methanolic extracts of *B. floccifera*. The antibacterial activity has been observed in the methanolic extracts of *B. floccifera* against the tested bacteria with varied activity. The maximum zone of inhibition was 28 mm for Bacillus cereus, 25 mm for Staphylococcus aureus, 15 mm for Escherichia coli, 13 mm for Proteus mirabilis, 7 mm for Klebsiella pneumonia. The other pathogens *viz.*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Serratia marcescens*, *Enterobacter* sp., *Enterococcus faecalis* and *Streptococcus pyogenes* showed the minimal inhibition only.

Table 1
Phytochemical studies on the methanolic flower extracts of *B. floccifera*.

Phyto-constituents	Occurence
Alkaloids	-
Phenol	+
Flavonoids	+
Saponins	+
Aminoacids	-
Quinones	-
Steroids, Phytosterols, Triterpenoidal Sapogenins	+
Tannins	+
Xanthoproteins	+
Carboxylic acid	-
Coumarins	+
Carbohydrates	+

+ - indicates the presence of the compound; - indicates the absence of the compound

Table 2
Antibacterial activity of the methanolic flower extracts of *B. floccifera*.

Microorganisms	*B. floccifera*	Amikacin
Escherichia coli	15	17
Klebsiella pneumoniae	07	18
Pseudomonas aeruginosa	02	25
Staphylococcus aureus	25	17
Bacillus cereus	28	40
Salmonella typhi	01	20
Serratia marcescens	03	18
Proteus mirabilis	13	19
Enterococcus faecalis	01	13
Streptococcus pyogenes	02	20

4. Discussion

Mohan *et al.*, [21] used chloroform, benzene and methanol as a solvent source. In the present study we used the methanol as solvent source for the extraction of the metabolites. Since the polarity of methanol is higher, most of the secondary metabolites of *B. floccifera* flower were dissolved. Out of 12 qualitative tests screened for the presence of secondary metabolites 8 showed positive results. Flavonoids have been referred to as nature's biological response modifiers because of strong experimental evidence of their inherent ability to modify the body's reaction to allergen, virus and

carcinogens. They show anti-allergic, anti-inflammatory, anti- microbial and anti-cancer activity [22, 23]. Tannins are known to possess general antimicrobial and antioxidant activities [24]. Recent reports show that tannins may have potential value as cytotoxic and antineoplastic agents [25]. Other compounds like saponins also have anti-fungal properties [26]. Saponins are a mild detergent used in intracellular histochemistry staining to allow antibody access to intracellular proteins. In medicine, it is used in hyper cholestrolaemia, hyperglycemia, antioxidant, anticancer, anti inflammatory and weight loss, etc. It is also known to have anti-fungal properties [27]. Saponins have been implicated as bioactive antibacterial agents of plants [28, 29]. Plant steroids are known to be important for their cardiotonic activities, possess insecticidal and anti- microbial properties. Plant derived natural products such as flavonoids, terpenoids and steroids etc have received considerable attention in recent years due to their diverse pharmacological properties including antioxidant and antitumor activity. Phenolic phytochemicals have antioxidative, antidiabetic, anticarcinogenic, antimicrobial, antiallergic, antimutagenic and anti-inflammtory [30, 31]. It suggests that the plants can be used as anti-microbial activity, anti-oxidant, anti-allergic, anti-inflammatory, antidiabetic, anti-carcinogenic, anti-cancer agents in the future. The methanol extracted solvents showed high degree (10/10 pathogens) of antibacterial activity against the selected pathogens with varied rate of inhibition. Klebsiella pneumoniae is an important cause of human infections and several diseases viz., urinary tract infections, noscomial infections, pneumonia, septicemias and soft tissue infections. The diseases caused by Klebsiella pneumoniae can result in death of patients who are immunodeficient. Streptococcus pyogenes causes many important human diseases, ranging from mild superficial skin infections to life-threatening systemic diseases. Infections typically begin in the throat or skin. The pathogen *S. typhi* is known to cause fever and food borne illness. Virulent strains of Escherichia coli can cause gastroenteritis, urinary tract infections and neonatal meningitis. In rare cases, virulent strains are also responsible for haemolytic uremicsyndrome, peritonitis, mastitis, septicaemia and gram negative pneumonia [32]. B. cereus is responsible for a minority of foodborne illnesses (2-5%), causing severe nausea, vomiting and diarrhea [33]. Serratia marcescens can cause infection in several sites, including the urinary tract, respiratory tract, wounds, and the eye, where it may cause conjunctivitis, keratitis, endophthalmitis and tear duct infections. It is also a rare cause of endocarditis and osteomyelitis, pneumonia and meningitis. Enterococcus faecalis can cause endocarditis, as well as bladder, prostate and epididymal infections; nervous system infections are less common. In the present study we observed the anti-bacterial activity of the methanolic extracts of *B. floccifera* against was 28 mm for Bacillus cereus, 25 mm for Staphylococcus aureus, 15 mm for Escherichia coli, 13 mm for Proteus mirabilis and 7 mm for Klebsiella pneumonia. The results of the present study suggest that the methanolic flower extracts of *B. floccifera* can be used to treat nausea, vomiting, diarrhea, urinary tract infections, noscomial infections, pneumonia, septicemias etc.

It is hoped that this study would lead to the establishment of some compounds that could be used to formulate new and more potent antimicrobial drugs of natural origin. Studies are in progress to further evaluate the mechanisms of action of *B. floccifera* methanolic flower extracts on some organisms associated with fish and human diseases. Further work will emphasize the isolation and characterization of active principles responsible for bio-efficacy and bioactivity.

Conflict of interest statement

We declare that we have no conflict of interest.

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