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Phytochemistry and antibacterial activity of *Solanum nigrum* against nosocomial bacterial pathogens

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ABSTRACT

The present study was focused the antibacterial activity and phytochemical analysis of *Solanum nigrum* leaf extract using disc diffusion method. The nosocomial bacterial pathogens are *Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli, Proteus mirabilis* and *Pseudomonas aeruginosa* were used in this study. The zone of inhibition was determined by disc diffusion method of *Solanum nigrum* extract showed highest activity of *Staphylococcus aureus*(19 mm) and showed least activity of *Pseudomonas aeruginosa* (11 mm)at the concentration of 100 μ l. The phytochemical analysis of *Solanum nigrum* indicates the presence of alkaloids, saponins, tannins, steroids,flavonoids and reducing sugars. The study concludes that the plant possesses novel compounds with significant antibacterial property to conflict pathogenic infections.

Keywords: *Solanum nigrum*, nosocomial pathogens, phytochemical activity, disc diffusion method and antibacterial activity.

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INTRODUCTION

Patients with non-infectious diseases who are admitted to the hospital are at a significant risk of contracting a nosocomial infection. According to reports, 10% of hospital patients get this infection while in the hospital [1]. In most cases, nosocomial infections originate in the respiratory and urinary tracts. The most frequent pathogenic bacteria that cause nosocomial infections are *Escherichia coli, Klebsilla pneumoniae*, *Haemophilus influenzae*, *Streptococcus pneumoniae* and *Proteus vulgaris*[2,3].

Traditional and modern medical systems both rely heavily on medicinal plants. Due to a number of side effects from the use of synthetic pharmaceuticals, antibiotics, and expensive costs, their usage has been expanded through numerous studies and applications. Because modern medicines and hospitals are not readily available in rural areas, people rely heavily on traditional medicine to treat their problems [4]. An effective approach in the discovery of new anti-infective agents from higher plants of phytochemical research is generally based on the ethnopharmacological information [5]. Plant materials remain an important resource to conflict serious diseases in the world. Traditional medicinal plants still play a significant role in the basic health needs of developing countries. The most important chemical bioactive constituents of plants are alkaloids, flavonoids, phenolic compounds, and tannins [6].

Solanum is one of the largest and extensively diverse genera of the family Solanaceae and it is well known in English and Tamil systems as "Black nightshade" and "Manathakkali" [7]. The leaves of Solanum nigrum are eaten as vegetables in most parts of the world wide the ripe seeds are also edible [8]. S. nigrum of leaves and fruits is chewed and swallowed to cure mouth ulcers [9]. The plant also possesses antimicrobial, antioxidant, cytotoxic properties, antiulcerogenic and hepatoprotective activity with a potentially herbal alternative that acts as an anti-cancer agent [10]. It has been extensively used in traditional treatment for various ailments such as pain, inflammation, and fever. It has been also extensively used in traditional medicine in India and other parts of the world to cure liver disorders, cough, asthma, wounds, ulcers, leprosy, skin diseases, haemorrhoids, dropsy, and inflammations. It has been used for the treatment of microbial and nonmicrobial diseases successfully in traditional medicine. Many of the plant materials are used in traditional medicine are readily available in rural areas and this system of medicine is relatively cheaper than modern medicine. As a result, many potent drugs have been purified from plants, including emetin, quinine, artemisin, and introduced to modern medical practice [11].

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Species in the *Solanum nigrum* exhibit considerable variation in genetically, both florally and vegetatively. In recent times, the use of herbal plants as a important source of novel compounds to combat microbial infections has gained prominence. The necessity to search for plant-based antimicrobials is increasing due to high cost, reduced efficacy and increased resistance to conventional medicine [12]. This research is important because it is aimed at establishing the specific medicinal value as well as the best species or variant of the *S. nigrum* for use as herbal medicine.

MATERIAL AND METHODS

Collection of Plant samples

Fresh plant samples of Solanum nigrum were collected from local market, Chidambaram, India.

Bacterial Strains

The test bacterial pathogens of Gram-positive bacteria *Staphylococcus aureus* (ATCC25923) *and Streptococcus pyogenes* (ATCC19615)and Gram- negative bacteria *Escherichia coli* (ATCC25922), *Proteus mirabilis* (ATCC12453) and *Pseudomonas aeruginosa* (ATCC27853). Bacteria were grown in Muller Hinton broth at 37°C for 24h and maintained on Muller Hinton agar slants at 4°C.

Preparation of the leaf extract

The leaves of *Solanum nigrum* plant leaf extract was collected and washed thoroughly with distilled water to removes the dust particles. The extract was prepared by using 20 g of washed dried finely cut leaves and stirred with 100 mlof sterile distilled water. The mixture was then boiled for 20 min until the colour of the aqueous solution changes from watery to light yellow colour. The leaf extract was filtered with Whatman No.1 filter paper and cooled at room temperature. And, the filtrate was stored at 4°C in order to use for further study.

Antibacterial assay

The antibacterial activity of *Solanum nigrum* were tested against gram-positive (*Staphylococcus aureus and Streptococcus pyogenes*) and gram-negative (*Escherichia coli, Proteus mirabilis*and *Pseudomonas aeruginosa*) bacteria by Kirby-Bauer disc diffusion method [13]. The bacterial suspension was swabbed on the Mueller Hinton Agar (MHA) plates by using sterile cotton swabs. Then 20µl of the plant extract was pipetted on a 5mm sterile paper disc, the solvent was allowed to evaporate and a disc was placed on the surface of the plate at aseptic conditions and the test plates were incubated for 24 hat 37°C. 25µg of Gentamycin was used as positive control.After the incubation period, antibacterial activity was measured as zone of inhibition was observed and interpreted.

Determination of the phytochemical composition

The extracts of leaves were subjected to phytochemical screening to qualitatively check the presence of phytochemicals in the extracts. The presence of the compounds was identified by color changes. And the results were reported as (+) for presence and (-) for absence.

Test for Saponins

The plant crude extract was mixed with 5ml of distilled water in a test tube and it was shaken vigorously. The stable foam indicates the presence of saponins.

Test for Phenols and Tannins

The plant crude extract was mixed with 2 ml of 5% solution of FeCl3. A blue-green coloration indicates the presence of phenols and tannins.

Test for Flavonoids

1 or 5 drops of concentrated hydrochloric acid (HCl) were added to the little amount of aqueous extract of the plant material. The development of red colour indicates the presence of flavonoids.

Test for Terpenoids

The plant crude extract was dissolved in 2ml of chloroform and evaporated to dryness. And 2ml of concentrated H2SO4 was added and heated for about 2 minutes. A greyish coloration indicates the presence of terpenoids.

Test for Alkaloids

2ml of plant crude extract was taken in a test tube and then 0.2 mL dilute HCl was included, followed by 1 mL of Meyer's reagent. A yellowish coloration indicates the alkaloid's presence.

Test for steroids

2 ml of extract, 2 ml of chloroform, and a concentration of H2SO4 were added. The presence of steroids indicates the red color production in the chloroform lower layer.

Test for Keller-kilani (Cardiac glycoside)

2 ml of plant crude extract was mixed with 2ml of glacial acetic acid containing 1-2 drops of 2% solution of FeCl3. The mixture was added to another test tube containing 2ml of concentrated H2SO4. The interphase of the brown ring indicates the presence of cardiac glycosides.

Test for Phlobatannins

 $2\,$ ml of plant sample and hydrochloric acid (1%) was added. Phlobatannins was confirmed on the formation of reddish-brown

Test for Quinones

A small amount of plant crude extract was treated with concentrated HCl and observed for the formation of a vallow color precipitate.

of a yellow color precipitate.

Reducing sugars

2 ml of plant crude extract and 5-8 drops of Fehling's solution were added and heated over a water bath and observed the brick red precipitate indicated the presence of reducing sugars.

RESULT AND DISCUSSION

The present study reveals that Solanum nigrum plant shows the presence of phytochemical constituents like alkaloids, steroids, saponins, terpenoids, tannins in aqueous extract as shown in (**Table1**)

S.NO	Phytochemicals	Result
1.	Alkaloids test	+
2.	Terpenoids test	-
3.	Quinones test	-
4.	Saponins test	+
5.	Steroids test	+
6.	Tannins test	+
7.	Reducing sugar test	+
8.	Kellerkillani test (Cardiac glycoside)	-
9.	Phenols test	-
10.	Phlobatannins test	-
11.	Flavonoids test	+

Table 1: Phytochemical screening of leaf extract of Solanum nigrum



+ Presence, - Negative **Figure 1:**Qualitativephytochemical analysis of *Solanum nigrum* leaf extract

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The results of the antibacterial activity showed that the plant extracts of *Solanum nigrum* showed the significant antibacterial property against *Staphylococcus aureus*, *Streptococcus pyogenes*, *Escherichia coli* and *Proteus mirabilis* and least significant activity against *Pseudomonas* aeruginosa. The aqueous extract of *Solanum nigrum* was found more potent against *Staphylococcus aureus*(19mm) and *Escherichia coli* (17mm) at the concentration of 100 mg/ml. Gentamycin used as positive control and Dimethylsulfoxide (DMSO) as negative control showed no activity against the tested bacterial strains. The antibacterial activity of plant extracts were evaluated by their zone of inhibition detailed results of the antibacterial potential of plant extracts are shown in **(Table 2)**.

	Concentration of leaf extract						
S.No	Bacterial strain	Zone of Inhibition (mm)					
		25µl	50µl	75µl	100µl	Positive	
						control	
1.	Escherichia coli	8	10	14	17	22	
2.	Staphylococcus aureus	11	13.5	15	19	25	
3.	Streptococcus pyogens	9	12	15	16	23	
4.	Pseudomonas aeruginosa	8	9	10	11	24	
5.	Proteus mirabilis	9	10	12	14	26	

Table2: Antibacterial activity of leaf extract of Solanum nigrum

Traditional healers used only water for the purpose of extraction but studies have proven the determining the pharmacological activity of a medicinal plant by using extracting solvent [14]. The plant may possess antimicrobial activity due to the presence of any of these phytoconstituents which may operate through varied mechanisms including the disruption of cell membrane, inhibition of cell wall formation, inactivation of microbial adhesins, suppression of enzymes or blocking of nucleic acid synthesis [15]. The biologically known compounds are active because they protect the plants against infection. The important sources of medicinal plants are potentially useful structures for the development of new chemotherapeutic agents. Phenolic compounds of phytochemicals have antioxidative, antidiabetic, anticarcinogenic, antimicrobial, antiallergic, antimutagenic and anti-inflammatory [16]. Tannins are known to possess general antimicrobial and antioxidant activities [17].

CONCLUSION

The present study demonstrates that *Solanum nigrum* contains good source of phytochemical constituents like alkaloids, saponins, tannins, flavonoids, steroids and reducing sugar. The antibacterial activity of *Solanum nigrum* was clearly shown the present study against the test organisms like *Staphylococcus aureus, Escherichia coli, Streptococcus pyogens, Pseudomonas aeruginosa* and *Proteus mirabilis.* The plant extract may contain potent antibacterial compounds, effective in the treatment of various bacterial infections. However, further investigation is needed to isolate the pure compound,

pharmacological studies as to use the promising compounds as an antimicrobic agents.

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