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SCREENING OF ANTIBACTERIAL ACTIVITY OF *SOLANUM MELONGENA* SEED EXTRACTS ON SELECTED HUMAN PATHOGENIC BACTERIA

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ABSTRACT

Antimicrobial efficiency of *Solanum melongena* seed extracts was examined using methanol, chloroform, acetone and petroleum ether, as solvents and tested against four human pathogenic bacteria such as *Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus* and *Proteus vulgaris* using Agar Well Diffusion Method. The phytochemical analysis of *Solanum melongena* seed was also made. The methanolic and acetone extract of *Solanum melongena* seed showed maximum inhibition against *Staphylococcus aureus* at all concentrations (75, 150 and 250 ug/ml) significant inhibition was proved at the higher concentration of petroleum ether extract while no inhibition was shown in the chloroform extract of Solanum *melongena* seed at any level of concentration against *Staphylococcus aureus*. The methanol extract of the *Solonum melongena* extract showed enormous inhibitory activity at all concentrations against *Escherichia coli* while the acetone and petroleum ether extracts of the same plant seed showed significant activity only at the higher concentration against *Escherichia coli*. The result of phytochemical analysis shows the presensce of reducing sugar, steroid, terpenoid and flavonoid in the seed extract of *Solanum melongena.*

KEYWORDS: ***Solanum melongena*, agar well diffusion method, antibacterial activity, phytochemical test**

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INTRODUCTION

Plants, the primary sources of medicine, have been playing a pivotal role in reducing health services around the globe (Thomson, 2010). About three quarters of the world's population relies on plants and their extracts for health care (Kunwar and Bussmann, 2008). A good number of our population depends largely on herbal remedies for the different types of diseases. It clearly indicates the important role of the individual plants in the health care system. (Amit subedi *et al.,* 2012) Plant-derived products contain a great diversity of phytochemicals such as phenolic acids, flavonoids, tannins, lignin and other small compounds (Cowan, 1999). Plant extracts have numerous health related effects such as antibacterial, antimutagenic, anticarcinogenic, antithrombotic and vasodilatory activities (Bidlack *et al.,* 2000). Organic solvents such as ethanol, acetone and methanol are often used to extract bioactive compounds (Eloff, 1998). Antimicrobial drugs have proved remarkably effective for the control of bacterial infections. The antimicrobial compounds from plants may inhibit bacterial growth by different mechanisms than those presently used as antimicrobials and may have a significant clinical value in the treatment of resistant microbial strains (Harborna, 1998). Flavonoids are a group of polyphenolic compounds found abundantly in the plant kingdom. An increased interest in the study of the possible health benefits of flavonoids and other polyphenolic compounds is shown in recent years owing to their potent antioxidant and free-radical scavenging activities (Bravo, 1998.,Heim *et al.,* 2002., Pier-Giorgio,2000.,Rice-Evans *et al.,* 1997.,Sealbert *et al.,* 2005.,Rose and Kasum,2002.,Rice Evans *et al.,* 1995) *Solanum melongena* (Egg plant) fruit popularly known as aubergine (UK), melanzana, garden egg, brinjal (India) (Singh *et al.,* 2009) is one of the most important vegetable crops grown all over the world. It nearly covers an area of 1.7 million hectare throughout the world. Egg plant contains a higher content of free reducing sugars, anthocyanin, phenols, glycoalkaloids and amide proteins. Bitterness in egg plant is due to the presence of glycoalkaloids. Egg plant is known to have some medicinal properties and is said to be good for diabetic patients. It has also been recommended as an excellent remedy for liver complaints (Sulunkhe and Kadam). Egg plants are best grown in sandy loam or silt loam soils with a pH of 5.5 to 6.8 at optimum temperature of 21-29^{o}C. Seeds are best sown at a temperature of 24-29^{o}C (Chen and Di, 1996). In this work, the antibacterial activity of the extracts of methanol, chloroform petroleum ether and acetone combined with *Solanum melongena* seed were comparatively studied against the human pathogenic bacteria, *Pseudomonas aeruginosa, Escherichia coli, Staphylococcus aureus* and *Proteus vulgaris*

MATERIALS AND METHODS

Preparation of plant extract

The seeds of Brinjal (*Solanum melongena*) were collected, washed thoroughly, air-dried under shade and finally milled to a coarse powder. The powdered material was obtained through soxhlet after 48 hours on using different solvents like methanol, chloroform, petroleum ether and acetone. All the extracts after evaporating them in vacuum under reduced pressure, were stored in sterile glass bottles at room temperature until screened.

Bacterial Strains

Pure cultures of human pathogenic bacterial strains, *Psuedomonas aeruginosa* (MTCC 741), *Escherichia coli* (MTCC 728), *Staphylococcus aureus* (MTCC 3160), and *Proteus vulgaris* (MTCC 7299) were bought from IMTECH, Chandigarh and the same bacterial strains were used for this study.

Antimicrobial test

Antibacterial activities of different extracts were evaluated by Agar well Diffusion Method (Murray *et al.,* 1995) later modified by Olurinale, 1996. Nutrient Agar (40 gm/L) plates were spread (sterile cotton swabs) with 8 hour old broth culture wells of 10 mm diameter and about 2 cm apart were made on each of these plates using sterile cork borer. Stock solution of plant extract was prepared at

a concentration of 1 mg/ml in the plant extract *viz* methanol, chloroform, petroleum ether and acetone. About 100 ml of different concentration of the plant extract was added through sterile syringe into the wells and allowed to diffuse at room temperature for 2 hours. Control experiment comprising inoculums without plant extract was set whereas the plates were incubated at 37^{o}C for 18-24 hours. The development of the zone was noticed. Triplicates were maintained and the experiment was repeated thrice. After the incubation period, the result was observed and the diameter of inhibition zone around each well was taken.

Photochemical Screening

The methanolic extract of the *Solanum melongena* seeds was separately subjected to preliminary phytochemical tests using standard methods while the Mayer's Hagner and Hagner's tests were carried out for alkaloids,Foam and hemolytic tests were used for saponins. Lieberman – Burchard and Salkowski for steroids / triterpenoids and Schinoda test was conducted for flavonoids. Then, $FeCl_3$, Lead acetate and Potassium dichromate test for tannins and phenolics and Molisch test for carbohydrates were done (Trease & Evens 1983)

RESULTS AND DISCUSSION

Plants are the important sources of potentially useful structures for the development of new chemotherapeutic agents.(Tona *et al.,* 1998). Phytochemicals derived from plant products serve as a prototype to develop less toxic and more effective medicines in controlling the growth of micro organism (Kelmanson *et al.,* 2000; Ahmed and Beg, 2001). These compounds have significant therapeutic application against human pathogens, including bacteria, fungi or virus. Numerous studies have been conducted by using the extracts of various plants, screening antimicrobial activity and discovering new antimicrobial compounds (Guleria and Kumar, 2006; Zakaria *et al.,* 2007). Therefore, medicinal plants play a prominent role and proffer a major contribution to pharmaceuticals neutralceuticals and food supplements. In the present investigation, the antibacterial effect of different extracts such as methanol, chloroform, acetone and petroleum ether of seeds from *Solanum melongena* were evaluated against the human pathogenic bacterial strains. The antimicrobial activity was determined by using agar well diffusion method was summarized in Table.1. Among the extractions assayed, the methanol extract of *Solanum melongena* seed gave maximum inhibition against *Staphylococcus aureus* and *Escherichia coli* at all concentrations (75, 150 and 250 mg/ml). It is quite obvious that the inhibition level gradually increases in accordance with the level of increase in concentration. However the same methanolic extract of *Solanum melongena* seed did not have any inhibitory activity against *Pseudomonas aeruginosa* and *Proteus vulgaris*. It is noteworthy to mention that Ghosh *et al.,* (2008) evaluated the antibacterial potentiality of hot aqueous and methanol solvent extracts of mature leaves of *Polyalshia longifolia* against six bacteria. On the other hand, Shirsat (2008) reported the anti phytopathogenic activity of crude and methanol extracts of leaves, stem, bark, seed and dry fruit of *Teminalia thoreii* against four phytopathogens. In the present study, the chloroform extracts of *Solanum melongena* seed did not show any antibacterial activity against the tested bacterial strains such as *Staphylococcus aureus, Pseudomonas aeruginosa, Proteus vulgaris* and *Escherichia coli* at any concentration. The acetone extract obtained from *Solanum melongena* seed exhibited a constant growth of activity against *Staphylococcus aureus* in accordance with the increase in the concentration. Surprisingly, an enormous amount of activity was found in the petroleum ether extract of *Solanum melongena* seed against *Staphylococcus aureus* at the higher concentration. Murugesan (2011) proved that a petroleum ether extract of plant *Menecylom umbellatum* Burm showed significant antimicrobial activity. Similarly the acetone and petroleum ether extracts of *Solanum melongena* seed showed significant activity against Escherichia coli at the higher concentration only.

Table 1
Antibacterial activity of Bringal (Solanum melongena seed)

Name of the bacterial strain	Different extract	Zone of inhibition (mm)		
		Extract concentration		
		75 µg/ml	150 µg/ml	250µg /ml
Staphylococcs aureus	Methanol	7±0.0	10±0.0	14±00
	Chloroform	0±0.0	0±0.0	0±0.0
	Acetone	5.6±05	7±1.5	16.±0.0
	Petroleum ether	0±0.0	8.3±0.5	10.6±0.57
Pseudomonas aeruginosa	Methanol	0±0.0	0±0.0	0±0.0
	Chloroform	0±0.0	0±0.0	0±0.0
	Acetone	0±0.0	0±0.0	0±0.0
	Petroleum ether	0±0.0	0±0.0	0±0.0
Proteius vulgaries	Methanol	0±0.0	0±0.0	0±0.0
	Chloroform	0±0.0	0±0.0	0±0.0
	Acetone	0±0.0	0±0.0	0±0.0
	Petroleum ether	0±0.0	0±0.0	0±0.0
Escherichia coli	Methanol	9.6±0.57	15±1.0	25±0
	Chloroform	0±0.0	0±0.0	0±0.0
	Acetone	0±0.0	0±0.0	9.3±0.26
	Petroleum ether	0±0.0	0±0.0	14±011

It was found that no inhibitory activity was found in any of the extracts (methanol, chloroform acetone, petroleum ether) with *Solanum melongena* seed against the bacterial strains *Pseudomanas aeruginosa* and *Proteus vulgaris*. (Dulger *et al.,* 1998) found that although four different extracts of a Macrofungus had antibacterial activity against *Bacillus subtilis* and some other Gram positive and Gram negative bacteria, they had not this activity against *E.coli*, *S.epidermidis* and *S.aureus.* The inhibitory activity of plant extract may vary according to the nature of active ingredients in the plant. Successful prediction of bioactive components from plant materials is largely dependent on the type of solvents used in the extraction procedure (Jayshree *et al.,* 2009). Phytochemical screening of *Solanum melongena* seed extracts (Table 2) revealed the presence of reducing sugar, steroid, terpenoid and flavonoid as the major active secondary metabolites. But, the Phytochemical constituents if used on other parts of the plant, the presence of secondary metabolites may vary accordingly. It is a fact that the presence of saponins and glycoalkaloids protects *Solanum melongena* plant from microbial pathogens. Quite contrary to the above work Paczkowski *et al.,* study proved the biosynthesis of steroidal, saporin and glycoalkaloids.

Table 2
Phytochemical Analysis

Samples	Test							
	Tannin	Phytotanin	Saponin	Reducing sugar	Alkaloids	Steroids	Terpenoids	Flavonoids
Brinjal	-	-	-	+	-	+	+	+

The present investigation concludes that the *Solanum melongena* seed contains potential antimicrobial components that may be of great use for the development of pharmaceutical industries to make use of them as a therapy against various diseases. The methanol, acetone and petroleum ether extracts of *Solanum melongena* possess significant inhibitory effect against *Staphylococcus aureus* and *Escherichia coli*. The present study may stand as a foreground experiment for future researches in which *Solanum melongena* seed may prove to be a major therapy in curing numberless chronicle diseases that affect the whole mankind for many years in various ways.

CONCLUSION

By performing the above work, it can be concluded that *Solanum melongena* seed extracts possess the antibacterial activities and the inhibition depends on the type of extract. The methanol and acetone extract of *Solanum melongena* seeds showed the higher antibacterial activity. Further studies should be undertaken in order to draw solid conclusion.

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