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Antibacterial activity of *Abutilon indicum Linn.*

Amit Kumar*, Navneet, Sanjay and Prabhat
Department of Botany & Microbiology, Gurukul Kangri University, Haridwar.
* Regional Drug testing laboratory, Chandigarh
Ministry of health and family welfare, Govt. of India.

Abstract

The *in vitro* antibacterial activity of leaves, stems, roots and seeds of *Abutilon indicum* Linn. have been investigated against human pathogenic bacteria i.e. *Klebseilla pneumoniae*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus mutans*, *Bacillus megnetherium* and *Escherichia coli* . The ethanolic extract showed the maximum antibacterial activity followed by petroleum ether and aqueous against all pathogens.

Introduction

Plants have a great potential for producing new drugs of great benefit to mankind. There are many approaches to the search for new biologically active principles. Many efforts have been done to new discover new antibacterial compounds from various kinds of sources such as soil, microorganisms, animals and plants, one of such resources is folk medicine and systematic screening of them may results in the discovery of novel effective compounds (Parekh and Chanda,, 2006).The microbes are slowly becomes resistant to drugs and some genetic changes take place in these microbes in course of time. The resistant strains are continuously appearing, imposing the need for a permanent search and development of new drugs, there is a need to develop an alternative to these drugs for the treatment of microbial diseases. Some medicinal herbs represent a rich source of antibacterial activity (Prabhat *et al*, 2005).In present study the *in vitro* antibacterial effect of different parts of *Abutilon indicum* Linn against *Klebsiella pneumoniae, S. aureus*, *B. megnetherium , Escherichia coli* , *S. mutans* and *S. epidermidis* have been studied.

Material and Methods

The different parts (leaves, stems, roots, seeds) of medicinal plant *Abutilon indicum* Linn. were collected from the local areas of Meerut and were identified by Botanical Survey of India (BSI), Dehradun. All parts of the plant were powdered by using grinder and extracted with petroleum ether, ethyl alcohol and water and the obtained extracts were dried by using vacuum rotatory evaporator under reduced pressure.

Test Organisms

Staphylococcus aureus MTCC 1144, *Staphylococcus epidermidis* MTCC 435, *Streptococcus mutans* MTCC 890, *Bacillus megnetherium* MTCC 453, *Escherichia coli* MTCC 433 and *Klebsiella pneumoniae* MTCC 109 were used.

Antimicrobial assay

Antimicrobial study was carried out by cup plate method (Prabhat *et al* 2005). The medium used for assay was Muller Hinton Agar media (Hi media No. 173). The fresh culture of each organism was prepared by inoculating a loopful growth from respective media.105 CFU/ml of culture was mixed in Muller Hinton agar. well of 6mm diameter were punched into agar with sterilized cork borer and each well was filled with 45

micro liter(100mg/ml) of plant extracts, solvents for blank and antibiotic (Ampicillin 100mg/ml) for positive control. The plates were incubated at 37 °C for 24 hrs .After incubation, a clear zone of inhibition was formed around the disc. Measure it with the help of zone reader. The average zone of inhibitions were noted. The zone includes the size of cup (6mm) and % of potency was calculated from the mean values as (Reyes chilpa *et al* 1997).

$$\text{Percentage of potency} = \frac{C - T}{C} \times 100$$

Where, C= positive control, T= Test

Results and Discussion

Results of antibacterial activities of the isolated materials obtained through petroleum ether, ethanol and water are summarized in Table 1-4. *Abutilon indicum* has an important place in the Indian system of medicine and the most parts of the plant are used therapeutically (Ahmad *et al* 1999). The different parts i.e. (leaves, stems, roots and seeds) were extracted in petroleum ether, ethyl alcohol and water. Most of the extracts showed prominent antibacterial activity against microorganisms i.e. *S. aureus*, *S. epidermidis*, *K. pneumoniae*, *B. megnetherium*, *S. mutans* and *E. coli*, at the concentration of 100mg/ml. as given in Table (1-4). The ethanolic extract showed the maximum activity followed by petroleum ether and water against all pathogens. The petroleum ether extract of leaves is highly active against *S. aureus* followed by , *S. epidermidis, B. megnetherium* and *E. coli*, as compared to other solvents. The percentage of potency of leaves extract against all bacteria in comparison to antibiotic was maximum in ethanol followed by petroleum ether and water.

The petroleum ether extract of stem is highly active against *S. aureus*, followed by *S. epidermidis, K. pneumoniae*, *E. coli, S. mutans* and *B. megnetherium* as compared to other solvents. The percentage of potency of stem extract in comparison to antibiotic was much in petroleum ether and followed by water and ethyl alcohol. The root extracts of plant *Abutilon indicum* is highly active against *S. epidermidis* followed by *K. pneumoniae, S. aureus, B. megnetherium* and *S .mutans* .The percentage of potency of petroleum ether extract of roots is maximum and followed by water and ethyl alcohol. The water extract of seeds of *A. indicum* is highly active against *K. pneumoniae* followed by *S. aureus* and *B. megnetherium*.

The crude extracts (100mg/ml) of each part were used for determination of their potency against pathogens as compared with antibiotic (100mg/ml).The extracts based on inhibition zone diameter has described as low (12-18 mm) moderate (19-22 mm) and strong activity (23-38 mm) (Ahmad *et al* , 1999). In our study all parts of plant showed low to moderate activity against pathogens. The ethanolic extract is highly effective against pathogens because more organic compounds were leached in this solvent. Although water is reported by the traditional healers and herbalists to be the most commonly used solvent for extracting the active compound due to its easy availability. Screening of medicinal plants to detect antimicrobial activity has clearly demonstrated that alcohol is better solvent as compared to water and petroleum ether.

The various parts of *Abutilon indicum* were tested for their antimicrobial activity against large number of bacteria (*S. aureus, S. epidermidis, K. pneumoniae, B. megnetherium, S .mutans* and *E. coli*. The water

extracts of leaves were devoid of any antibacterial activity against *K. pneumoniae*, *E. coli* and *B. cereus* etc., but in our study it was active against both gram positive and gram negative bacteria. The above results showed antibacterial activity of this plant and are correlated with the traditional uses of this plant in ayurvedic system of medicine. Further work is required before they can be recommended as therapeutic agent. Our result shows that, the petroleum ether extract is highly active against *S .aureus, B. megnetherium* and *E. coli.* The chemical analysis of petroleum ether extract from roots of the plant showed the presence of caprylic, palmitic, myristic and oleic acid. Geta *et. al.* 1983 also reported that essential oils obtained from *A. indicum* exhibited antibacterial activity against *B. subtilis, P. vulgaris* while it is inactive against *S. pullorum, S. typhimurium* and *Klebsiella spp.*

Aqueous extract of the plant parts i.e. leaves stems, roots and seeds showed antibacterial activity against *S. epidermidis, B. megnetherium* followed by *S. aureus, K. pneumoniae, S .mutans* and *E. coli.* The water extract contain carbohydrates, amino acids, saponins and flavanoids. They have been found *in vitro* to be effective antimicrobial substances. This activity is probably due to their ability to complex with extracellular and soluble proteins and to complex with bacterial cell wall for quinones. More liphophilic flavanoids may also disrupt microbial membranes (Tsuchiya *et al.* 1996). Naqvi *et al.* 1991 found that aqueous extract of the plant at concentration of 5 to 10 mg/ml showed activity against *S. aureus, S. viridans, C. diphtheria E. coli, S. typhi, S. paratyphi A* and *B., S. flexneri* .Valsraj *et al.* (1997) reported that 80% of alcoholic extracts of the roots of *A. indicum* showed no activity against *E. coli, P. aeruginosa, B. subtilis* and *S. aureus.* In our study ethyl alcohol extract showed good activity against *E. coli* and *S. aureus*. The root of the plant was extracted in 95% ethyl alcohol and antimicrobial studies were carried out by cup plate method. The gallic acid was present during chemical analysis of the roots. Sato *et al.* 1995 reported that gallic acid is effective against *S. aureus*.

Mehta *et al*. 1997 while studying ethanolic and acetone extract of the roots exhibited significant antibacterial activity against *E. coli, Proteus* and *P. aeruginosa*. The hexane extract was active against *P .aeruginosa* and *S. aureus.* The chloroform and ethyl acetate extract showed good activity against most of the bacteria. Most of the Ayurvedic practioners use aqueous extracts. This substantiate the view that aqueous extract contain most of the polar and nonpolar compounds i.e. flavonoids, saponins, sugars etc and thus the aqueous extract is antibacterial in action. So, efficacy and toxicity studies remains to be tested before further exploitation of the drug for human treatment.

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Table1 : The percentage of potency of plant *Abution indicum* leaves extract and antibiotic (ampicillin) against pathogens

S. No	Pathogens	Zone of inhibition (Antibiotic 100 mg/ml/.)			Zone of inhibition (Extra 100 mg/ml.)			% of potency		
		P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water
1	*S. aureus*	16	18	19	No Zone	17	16	-	5.55	15.78
2	*S. epidermidis*	17	19	18	9	14	16	47.05	26.31	11.11
3	*K. pneumoniae*	15	17	17	No Zone	13	-	-	23.52	-
4	*B. megnetherium*	16	19	20	10	12	15	37.50	36.84	25.00
5	*S. mutans*	15	18	20	No Zone	12	14	-	33.33	30.00
6	*E. coli*	15	17	20	10	11	13	33.33	35.29	35.00

Table2 : The percentage of potency of plant *Abution indicum* stem extract and antibiotic (ampicillin) against pathogens

S. No	Pathogens	Zone of inhibition (Antibiotic 100 mg/ml/.)			Zone of inhibition (Extra 100 mg/ml.)			% of potency		
		P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water
1	*S. aureus*	16	18	19	9	13	14	43.75	27.77	26.31
2	*S. epidermidis*	17	19	18	10	14	13	41.17	26.31	27.77
3	*K. pneumoniae*	15	17	17	10	13	12	33.33	23.52	29.41
4	*B. megnetherium*	16	19	20	11	12	13	31.25	36.84	35.00
5	*S. mutans*	15	18	20	10	11	11	33.33	38.88	45.00
6	*E. coli*	15	17	20	10	12	12	33.33	29.41	40.00

Table3 : The percentage of potency of plant *Abution indicum* roots extract and antibiotic (ampicillin) against pathogens

S. No	Pathogens	Zone of inhibition (Antibiotic 100 mg/ml/.)			Zone of inhibition (Extra 100 mg/ml.)			% of potency		
		P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water
1	*S. aureus*	16	18	19	14	15	13	12.50	16.66	31.57
2	*S. epidermidis*	17	19	18	13	17	16	23.52	10.52	11.11
3	*K. pneumoniae*	15	17	17	11	14	13	26.66	17.64	23.52
4	*B. megnetherium*	16	19	20	10	13	15	37.50	31.57	25.00
5	*S. mutans*	15	18	20	11	12	14	26.66	33.33	30.00
6	*E. coli*	15	17	20	10	12	13	33.33	29.41	35.00

Table4 : The percentage of potency of plant *Abution indicum* seeds extract and antibiotic (ampicillin) against pathogens

S. No	Pathogens	Zone of inhibition (Antibiotic 100 mg/ml/.)			Zone of inhibition (Extra 100 mg/ml.)			% of potency		
		P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water	P. Ether	E. Alcohol	Water
1	*S. aureus*	16	18	19	11	14	13	31.25	22.22	31.57
2	*S. epidermidis*	17	19	18	10	15	13	41.17	21.05	27.77
3	*K. pneumoniae*	15	17	17	9	13	15	40.00	23.52	11.76
4	*B. megnetherium*	16	19	20	10	12	13	37.50	36.84	30.00
5	*S. mutans*	15	18	20	10	11	12	33.33	38.88	40.00
6	*E. coli*	15	17	20	11	12	12	26.66	29.41	40.00

*Zone of inhibition mm