

“Antimicrobial Investigation of *Datura stramonium* Leaf Extract against different Microorganisms.”

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Abstract: An extract of *Datura stramonium* was screened for phytochemical analysis and antimicrobial assay against different bacterial species. The solvent used for extract preparation was methanol. Phytochemical analysis of leaf extract shows presence of Alkaloids, phenols, carbohydrates, glycosides, Terpenoids, steroids and proteins. Agar well diffusion method was used for study of antimicrobial activity of leaf extract. Standard antibiotic (Azithromycin) and methanol were used as a positive and negative respectively. Leaf extract of *Datura stramonium* shows strong antimicrobial activity against bacterial species like *Bacillus thuringiensis*, *Pseudomonas aeruginosa*, *Agrobacterium tumefaciens* and *Klebsiella pneumoniae*.

Keywords: *Datura stramonium*, Antimicrobial activity, phytochemical analysis, Agar well diffusion, Azithromycin

I. Introduction

Datura stramonium is also known as thorn apple, Jamestown weed or devil's trumpet this weed belongs to family Solanaceae. *Datura stramonium* produces flowers of white to creamy or violet colour. Generally flower is of 2.5 to 3.5 in. long. They rarely open completely. Almost all plant parts of the *Datura stramonium* are toxic. (Radford et al., 1964). In India this plant found anywhere from roadside to farm as a weed. In little measures it was used in Ayurveda medicinal plant. (Devi et al., 2011)

II. Materials And Method

2.1 Plant material-

Fresh leaves of *Datura stramonium* were collected from Babhaleshwar farm i.e Agricultural land of K. K. Wagh College of Agricultural Biotechnology, Nashik, India. These fresh leaves were washed thoroughly with tap water and then by distilled water and then shade dried for 4-5 days. The dried leaves were powdered with the help of mixer and grinder and 10 gm of powder used for the extract preparation.

2.2 Preparation of Extract-

For the preparation of plant extract 20 gm of leaf powder was extracted in a Soxhlet Apparatus using 200 ml of methanol solvent. The extract were concentrated using rotary evaporator. The extract obtained were weighed and kept at 4°C. 15 mg of solvent residue was dissolved in 1 ml of DMSO (5%) as a solvent and were used as the test extracts for antimicrobial activity. (Fazal et al., 2011)

2.3 Phytochemical analysis-

The phytochemical analysis was carried out on the methanol extract using standard procedures to identify the phytochemical constituents. (Yadav and Munin, 2011)

- a. **Alkaloids-** 0.5 g of each sample were dissolved with 5 ml of 2 N HCl and filtered. Filtrate was treated with Dragendroff's reagent. Formation of red precipitate indicates the presence of alkaloid.
- b. **Phenols-** Extract was treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenols.
- c. **Carbohydrates-** To 1 ml of the filtrate, 5 ml of Benedict's reagent were added. The mixture was heated. Appearance of red precipitate indicated the presence of reducing sugar. (Harborne, 1984)
- d. **Glycosides-** 0.5 g of each extract was stirred with 10 ml of boiling distilled water. This was filtered and 2 ml of the filtrate hydrolyzed with a few drops of concentrated HCL and the solution rendered alkaline with a 5 drops of ammonia solution. 5 drops of this solution was added to 2 ml of Benedict's qualitative reagent and boiled. Appearance of reddish brown precipitate showed the presence of glycosides.
- e. **Terpenoids-** 5 ml of extract was mixed in 2 ml of chloroform and concentrated sulphuric acid (3ml) was carefully added to form a layer. A reddish brown colour formation shows positive results for the presence of terpenoids. (Kumar et al., 2007)

- f. **Steroids**-The extract was mixed with 2 ml of chloroform and concentrated sulphuric acid was added sidewise. A red colour produced in the lower chloroform layer indicated the presence of steroids.
- g. **Proteins (Millon's test)**- Take hot nitric acid and add the extract 2-4ml then add ammonia, if orange yellow color are observe then protein are present.

2.4 Test Microorganism-

All these test bacterial species were procured from National Chemical Laboratory (NCL), Pune (Maharashtra, India).

2.5 Antimicrobial Assay:-

Antimicrobial activity of solvent extract methanol was determined by Well Diffusion Method on Nutrient Agar medium. (Satdive et al., 2012)

A well with diameter 5mm was made using sterile cork borer and inoculums of bacteria were spread on the solid plates with a sterile swab moistened with the bacterial suspension. Then 50 μ g each of solvent extracts were poured in the wells of the inoculated plates. The plates were incubated for 24 hrs at 37⁰C and zone of inhibition if any around the wells were measured in mm.

III. Result

After extraction of Datura stramonium extract in Soxhlet using methanol as a solvent, extract was subjected to various quantitative phytochemical analysis result of phytochemical analysis is summarized in Table 1. The zone of inhibition produced by Datura stramonium leaf extract against microorganisms is observed and noted in Table 2. Standard antibiotic (Azithromycin) was used as a positive control and Methanol was used as a negative control.

Table (1): Phytochemical analysis of methanol extract of Datura stramonium

Sr. No.	Phytochemicals	Datura stramonium
1	Alkaloids	+
2	Phenols	+
3	Carbohydrates	+
4	Glycosides	+
5	Terpenoids	+
6	Steroids	+
7	Proteins	+

Table (2): Antimicrobial activity of methanol extract of Datura stramonium against different bacterial species.

Sr. No.	Bacterial cultures	Datura stramonium (in mm)	Azithromycin (in mm)
1	Bacillus thuringiensis,	16	19
2	Pseudomonas aeruginosa,	19	17
3	Agrobacterium tumefaciens	18	20
4	Klebsiella pneumoniae	19	20

IV. Discussion

In the present study, we have carried out the phytochemical analysis of methanol extract of Datura stramonium leaves. Alkaloids, Phenols, Carbohydrates, glycosides, Terpenoids, Steroids and proteins are present in extracts all these biochemical compounds are biologically active compounds.

In antimicrobial study we have found that Datura stramonium leaf extract shows antimicrobial activity against microorganisms Bacillus thuringiensis, Pseudomonas aeruginosa, Agrobacterium tumefaciens and Klebsiella pneumoniae by forming zone of inhibition of 16mm, 19mm, 18mm and 19mm respectively. From this study we can conclude that Datura stramonium has antimicrobial activity.

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