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Pharmacognostic And Preliminary Phytochemical Investigation Of Cassia Auriculata (L.,) Leaves

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Abstract
Leaves of plant Cassia Auriculata L. are reported to possess medicinal values in traditional leaves of medicine. The present investigation deals with preliminary phytochemical investigation of leaves of Cassia Auriculata L. which includes physicochemical parameters like ash values, extractive values, and moisture content. The total ash, acid-insoluble ash, water-soluble ash values, and sulfated ash were observed to be 5.04%, 2.96%, 2.13%, and 0.56% respectively. Alcohol-soluble and water-soluble extractive values of leaves were observed to be 2.73%, and 5.51% respectively. Phytochemical investigation of n-hexane, chloroform, and ethanol extract revealed the presence of glycosides, tannins, terpenoids, steroids, carbohydrates, alkaloids, saponins, and proteins. The main aim of the present investigation is to study the Pharmacognostic characteristics and phytochemical standard of leaves of Cassia Auriculata L. which could be used to prepare a monograph for proper plant identification. The antibacterial and antifungal activities of solvent extracts of Cassia Auriculata L. were tested against one gram positive, one-gram negative human pathogenic bacteria and one fungi respectively. All extracts showed broad spectrum of inhibition by showing antibacterial effect of both bacterial stains. The tested bacterial strains were S. aureus, E. coli and fungal strains was C. albicans. The antimicrobial activity of these extracts is due to the presence of secondary metabolites. Hence these plants can be used to discover bioactive natural products that may serve as leads in development of new pharmaceuticals research.
Keywords: *- Cassia Auriculata L., Physicochemical, Phytochemical analysis.*

INTRODUCTION

Antibiotics are one of our most important weapons in fighting bacterial infections. However, from the past few decades these health benefits are under threat many antibiotics become less effective against certain illness not only because many of them produce toxic reactions. In many developing countries, traditional medicine is one of the primary health care system. Cassia Auriculata L. commonly called as Aval is an annual herb found in throughout India and also in many tropical countries. The leaves are tasty and sour. This plant has been reported to exhibit antibacterial[1], hypoglycaemic and microbicidal activities. The shrub is particularly renowned for its vibrant yellow flowers, which are used to treat skin disorders and body odour[2]. Cassia auriculata L. has been reported to possess antidiabetic, antioxidant, antibacterial, anti-inflammatory, and other medicinal properties, making it a valuable plant in traditional medicine[3,4]. In continuation of the work of phytochemical studies of various plants, we are presenting this paper on Cassia Auriculata L.

MATERIAL AND METHODS

Plant Material Collection and Authentication

The Leaves of plant Cassia auriculata L. were collected from the village Kawathi of Dhule tehsil in Dhule district (M.S.). The specimens of plants were authenticated by Dr. D. G. Jadhav, Head, Research Centre of Botany, M.S.G. Art's, Commerce and Science College, Malegaon Camp, Nashik (M.S.). The dried uniform Leaves powder was used for the extraction of constituents of the plant, determination of ash values, extractive values and phytochemical investigation.

Drying and pulverization

Leaves of Cassia Auriculata L. were shade-dried and pulverized and stored in an air-tight container for future use.

Extraction of powdered Leaves[5]

The powdered Leaves were successively extracted by cold maceration process using organic solvents like ethanol, n-hexane, and chloroform. All the extracts were evaporated to dryness and stored for future use.

PHARMACOGNOSTIC STUDIES

Physicochemical Investigation

The moisture content, total ash, water-soluble ash, acid-insoluble ash, sulfated ash, alcohol and water-soluble extractive values were determined as part of its physicochemical parameters[6,7].

Phytochemical Investigation Ethanol, n-hexane, and chloroform extracts were subjected to phytochemical analysis for the presence of various secondary phytoconstituents using standard chemical tests[7, 8, 9].

Test microorganisms and growth media

S. aureus (NCIM 2079), E. coli (NCIM 2169) and fungal strain C. albicans (NCIM 3471) were chosen based upon their clinical and pharmacological importance (Mc Cracken et al; 1983). The bacterial strains obtained from NCIM Pune were used for evaluating antimicrobial activity. The bacterial and fungal stock cultures were incubated for 24 Hrs. at 37°C on Nutrient Agar and MGYP respectively following refrigeration storage at 4°C. The bacterial strains were grown in Muller Hinton agar at 37°C whereas the yeast were grown in MGYP respectively at 28°C. The stock cultures were maintained at 4°C.

Antimicrobial activity

In vitro antibacterial and antifungal activity were examined for ethanol, n-Hexane, Chloroform and water extracts. Antibacterial and antifungal activities of these extract against two pathogenic bacteria and one pathogenic fungi were investigated by the Agar Disk Diffusion method. All the extracts were screened for their antibacterial and antifungal activities against the S. aureus, E. coli and fungi strain C. albicans. The dilutions of C. Auriculata L. extracts and standard drugs were prepared in double distilled water using nutrient agar tubes. Muller Hinton sterile agar plates were seeded with bacterial strains (1 x 108 bacteria/ ml) and allowed to stay at 37°C for 3 hrs. Control experiments were carried out under similar condition by using Chloramphenicol for antibacterial activity and Nystatin for antifungal activity as standard drugs. All of the plates were incubated at 37°C for 18 to 24 hrs for bacteria and at 28°C for 48 to 96 hrs for fungi. The zones of growth inhibition around the disks were measured after 18 to 24 hrs of incubation at 37°C for bacteria and 48 to 96 h for fungi at 28°C, respectively. The sensitivity of the microorganism species to the plant extracts was determined by measuring the sizes of inhibitory zones (including the diameter of disk) on the agar surface around the disks.

RESULT AND DISCUSSION

Physical appearance, color and odor of different extracts were recorded in (Table 1).

Table 1: Shows characteristics of Cassia Auriculata L. extracts.

Sr. No.	Extract	Physical Appearance	Color	Odor
1	Ethanol	Semi-Solid mass	Dark Green	Pungent Aromatic
2	n- hexane	Syrupy mass	Light Green	Aromatic
3	Chloroform	Semi-Solid mass	Dark Green	Aromatic

The physical constants evaluation of drugs is an important parameter in detecting adulteration or improper handling of drugs. The total ash value is important in the evaluation of purity of drugs i.e., the presence or absence of foreign inorganic matter. The ash values, extractive values, and moisture content of Leaves were determined, and the results are shown in (Table – 2).

Table 2: Shows physicochemical parameters of Cassia Auriculata L. Leaves.

Sr.No.	Parameters	Values (%) w/w
1	Loss on drying	
	Ash values:	2.96%
	Total ash	5.04%
2	Acid insoluble ash	2.43%
	Water soluble ash	2.13%
	Sulphated ash	0.56%
3	Extractive values:	5.51%
	Water soluble extractives	2.73%

Alcohol soluble extractives

Phytochemical tests for the presence of secondary phytoconstituents showed the following results (Table 3); furthermore, the phytochemical screening of the extracts revealed the presence of various secondary phytoconstituents, and the results are summarized in Table 3.

Table 3: Show preliminary phytochemical screening of Cassia Auriculata L. Leaves powder.

Sr. No.	Phytoconstituents	Ethanol	n-Hexane	Chloroform
1	Alkaloids	_	_	_
2	Carbohydrates	+	+	+
3	Glycosides	+	+	+
4	Flavonoids	+	+	+
5	Phenol& Tannins	+	+	+
6	Steroids	_	_	_
7	Terpenoids	+	+	+
8	Saponins	_	_	_
9	Proteins	+	+	+
10	Amino Acids	+	+	+

The anti-microbial activity of all extracts of C. Auriculata L. were studied with concentration 100 µg/ml against two pathogenic bacterial strains and one fungal strain. Antibacterial and antifungal potential of extracts assessed in terms of zone of inhibition of bacterial growth. The results of antimicrobial activities are presented in Table 4-5. The growth inhibition zone measured range from 08-12 mm for sensitive bacteria and ranged from 08-10 for fungal strains.

The results showed that C. Auriculata L. leaves extracts were found to be effective against all the microbes tested.

Table 4: Antibacterial activity of extracts of C. Auriculata L. against bacterial test organism.

Microorganism	Zone Of Inhibition in mm Concentration in 100 µg/ml			
	n-Hexane Extract	Chloroform Extract	Ethanol Extract	Chloramphenicol Standard
E. coli	12.45	11.21	10.56	17.78
S. aureus	9.33	12.74	~	21.55
B. subtilis	10.32	11.14	3.87	20.25
P. vulgaris	8.50	10.85	8.24	16.49

Table 5: Antifungal activity of extracts of C. Auriculata L. against bacterial test Organism.

Microorganism	Zone Of Inhibition in mm Concentration in 100 µg/ml			
	Ethanol Extract	n-Hexane Extract	Chloroform Extract	Nystatin Standard
C. albicans	~	~	~	24.55
A. niger	~	~	~	20.20

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