ISSN: 0976-7126

INTERNATIONAL JOURNAL OF PHARMACY & LIFE SCIENCES

Phytochemical screening and antifungal activity of stem of *Argyreia speciosa* Linn. F.

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Abstract

Argyreia speciosa is a potent medicinal plant in the Indian systems of medicine. Traditionally it is used as the treatment of various diseases and disorder of human. In the present study the aqueous, ethanolic, chloroform and petroleum ether extract of the *A. speciosa* were studied for their antifungal activity against *Candila albicans*. It was observed that the ethanolic extract and petroleum ether extract showed significant activity whereas aqueous extract showed very less activity and chloroform extract did not showed any activity against the tested fungal strain.

Key-Words: *Argyreia speciosa*, Phytochemical screening, Stem

Introduction

Plants have been used in virtually all culture as a source of food, clothes and shelter. Beside, these they provide timber, fuel, dye, gum, resin, medicine etc. to us and have very significant role in human civilization. The dependence of tribal and rural people on plant based material is increasing day by day. Medicinal plants have always been the principle source of medicine in India since ancient time and presently they are becoming popular throughout the developed countries. They also play an important role in the life of tribal and rural people, particularly in remote part of developing countries. Obviously, these plants help in alleviating human suffering. These plants are being integrated to the field of foods as additives, beverages and cosmetics.[1-2]

Argyreia speciosa (Convolvulaceae), commonly known as Suruudrashok in Hindi is a woody climber and has been used as a 'rasayana' drug in the traditional Ayurvedic system of medicine. Traditionally the stem of this plant have been useful in stomach complaints, sores on foot, small pox, syphilis, dysentery and diarrhoea.[3] It is found throughout India, up to an altitude of 300m, viz., Assam, Bengal, Puri district of Orissa, Dehra Dun, cultivated in Rajasthan, Konkan, Deccan, Mysore. It is a large climber; stem stout, white-tomentose.

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Leaves are 7.5-30.0 cm. in diameter, acute, ovate, glabrous above, persistently white-tomentose beneath, base cordate; petioles 5-15 cm, long, white-tomentose. Flowers in subcapitate cymes; penduncles 7.5-15 cm, long, stout, white-tomentose; bracts large, ovate-lanceolate with a long acumen, thin, veined, pubescent outside, glabrous inside, deciduous the outer sometimes 5 cm, long; pedicels very short often almost 0, white-tomentose. Calyx white tomentose outside; corolla 5-6.3 cm, long, tubularin fundibuliform, the bands silky pubescent outside, tube somewhat inflated, white pubescent outside, rose purple and glabrous inside. Ovary glabrous. Fruit glabrous, 2.0 cm in diameter, apiculate[4]. The aim of the present study is to detect the phytochemical and investigate the antifungal spectrum from natural resources and to support the traditional uses of *Argyreia speciosa*.

Material and methods

Selection of plant

Argyreia speciosa Linn. F. belongs to family Convulvulaceae is medicinally important plant, commonly grown in some parts of our country and used in the treatment of various disease and disorders of human ailments. Therefore, the plant was selected for present investigation.

Collection of plant material

The stem of the selected plant was collected from the Medicinal gardens of UIPS, Ujjain (M.P.)

Extraction of Plant Material

The extraction of plant material (stem) was carried out by cold maceration.[5]

Aqueous extract

10 gm dried stem powder was taken and dissolved in 250 ml liter of water in 500ml beaker and left for 48 hours with frequent shaking, after that it was filtered using muslin cloth and the filtrate was collected and concentrated.

Chloroform extract

10 gm dried stem powder was taken and dissolved in 250 ml chloroform in 500ml beaker and left for 48 hours with frequent shaking, after that it was filtered using muslin cloth and the filtrate was collected and concentrated.

Petroleum ether extract

10 gm dried stem powder was taken and dissolved in 250 ml petroleum ether in 500ml beaker and left for 48 hours with frequent shaking, after that it was filtered using muslin cloth and the filtrate was collected and concentrated.

Ethanolic extract

10 gm dried stem powder was taken and dissolved in 250 ml ethanol in 500ml beaker and left for 48 hours with frequent shaking, after that it was filtered using muslin cloth and the filtrate was collected and concentrated.

Preliminary Phytochemical screening

The various extracts obtained after maceration was subjected to various phytochemical screening as per the standard procedure to reveal various active phytoconstituents.[5]

Anti-fungal activity[6]

Collection of micro-organism

Fungal strain *Candila* was obtained from RD Gardi Medical College, Ujjian, M.P.

Drug entrapped disc

Whatmann filter paper was pieces into small disc 1 quarter inch diameter. Dilutions were applied to autoclave filter paper disc using micro pipette with sterile pipette tip.

Cultured media

Culture media i.e., Saburauds agar media was prepared. The composition was mentioned in table 1.

Table 1: Composition of Saburauds agar media

Ingredients	Quantity prescribed	Quantity taken
Dextrose	40 g	10 g
Peptone	10 g	2.5 g
Agar	20 g	5 g
Dist water	1000 ml	250

Nutrient media

The nutrient media was prepared. 5 gm of Agar was dissolved in 250 ml of water, then 10 gm dextrose and 2.5 gm peptone was added in above solution with continuous stirring. The media was heated to dissolve the agar and formed a clear liquid. Then pH was adjusted to 5.4. The media was sterilized at 15 lb pressure and 115^0C for 15 minutes in an autoclave.

Dilution

100 mg of drug was dissolved in 100 ml of distilled water to prepared stock solution (1000 ug/ml). From the above solution, 10 ml was taken into the volumetric flask and diluted up to 100 ml distilled water to prepared sub stock solution. (100ug/ml). From the above solution the adequate solution concentration 20, 40, 60, 80 ug/ml was prepared. In the same way standard drug concentration 60 ug/ml was prepared.

Application of disc

All the disc of sample, standard, control with different dilution was placed on to the incubated plate with the help of flame sterilized forcep. After application of disc, lid of petri plate was closed. Petri plates were incubated for 72 hours at 37^0c.

Zone of inhibition

After incubation the plates were inspected to identify zone of inhibition. The diameter of zone of inhibition of each compound and the diameter of disc of different concentration was recorded with using the formula.

Zone of inhibition – Diameter of sample / Standard / control - Diameter of disc.

Results and discussion

Argyrieia speciosa Linn. F. is an indigenous herb which is chosen for the present investigation study. The plant belongs to the family Convulvulaceae. The scanty availability of information on this plant facilitates the study on it. This attempt was made to study the Extraction, preliminary phytochemical investigation and antifungal activity of plant.

The stem was dried under shade and was made to coarse powder. This powder was subjected to extraction using different solvents. Extractive values of various extracts were determined and the values indicate the presence of considerable amount of constituents which are soluble in them. The percentage yield of aqueous extract was found to be maximum i.e., 3.75 % w/w, followed by ethanolic extract 3.66 % w/w, petroleum ether 2.50 % w/w and chloroform extract 1.56 % w/w. The results are mentioned in Table 2.

The various extract of the plant were subjected to phytochemical screening which reveal the presence of various pharmacological active components.

The different concentrations (20, 40, 60, 80 ug/ml) of extracts were tested for antifungal activity. The most effective concentration was found to be of Ethanolic extract because it gave the maximum Zone of inhibition as compared to other three extract and is found to be optimum as compared to standard drug Fluconazole.

The antifungal action of constituents obtained from various extract of stem of *A. speciosa* on fungi (*Candida*) was studied using four different concentrations (20, 40, 60 & 80 ug/ml). On the basis of findings, the effect produced by extract was comparable to that of fluconazole, antifungal drug. Ethanolic extract showed the potent antifungal activity as compared to other extract and the Zone of inhibition of test drug is compared to standard. However the Zone of inhibition of test drug was increased with the increase in concentration. Thus, these studies provided a scientific support to the selected medicinal plants which claims its folk lore use in medicine.

Acknowledgment

The authors are thankful to the Management Ujjain Institute of Pharmaceutical Sciences for providing the necessary facilities in the college. Special thanks to Department of Microbiology, RD Gardi Medical College, Ujjain for providing the Fungal Strains.

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Table 2: Percentage of extract obtained

S/No.	Parameters	Estimated percentage(w/w)	Color	Nature
1.	Aqueous Extract	3.75	Green	Gummy
2.	Ethanolic Extract	3.66	Light Green	Powder
3.	Pet. Ether Extract	2.50	Dark green	Powder
4.	Chloroform Extract	1.56	Green	Powder

Table 3: Preliminary phytochemical screening of extracts

S/No.	Constituents	Test	Aqueous extract	Ethanolic extract	Chloroform extract	Petroleum ether extract
1.	Alkaloids	Mayer's test	-	-	-	-
		Dragendroff' test	+	+	-	-
		Hager's test	-	-	-	-
		Wagner's test	-	-	-	-
2.	Carbohydrates	Molisch's test	+	+	-	-
		Fehling's test	-	-	-	+
3.	Glycosides	Brontrager's test	+	-	-	-
		Legal's test	-	-	-	-

ISSN: 0976-7126

		Spot test	+	+	-	+
4.	Fixed oil and fats	Soap formation test	+	-	-	-
		$FeCl_3$	-	-	-	-
		Vanillin HCl	-	-	-	-
5.	Tannins	Alkaline reagent	-	-	-	-
		Million's test	+	-	-	-
		Ninhydrin test	+	+	-	+
6.	Protein and amino acid	Biuret test	-	-	-	-
		With NaOH	+	+	-	+
7.	Flavanoids	With H_2SO_4	-	+	-	-
		Libermann's Burchard test	-	+	-	-
8.	Steroids and triterpenoids	Salkowski's test	+	-	-	-
9.	Mucilage and gum	With 90% alcohol	-	-	-	-
10.	Waxes	With alc. KOH	-	-	-	-

Abbr.: +=Present; - =Absent

Table 4: Zone of inhibition of different extract of *Agryreia speciosa* Linn.

Strain	***Candila albicans***			
Concentration (ug/ml)	**20**	**40**	**60**	**80**
Sample				
Aqueous Extract	-	-	5	7
Ethanolic Extract	-	9	11	14
Chloroform Extract	-	-	-	-
Pet. Ether Extract	-	-	4	9
Standard Drug (ug/ml)	16 (F)			
Control	-			