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PHARMACOGNOSTIC, PHYTOCHEMICAL, ANTIOXIDANT AND ANTI-INFLAMMATORY STUDIES ON THE LEAVES OF *CHASSALIA CURVIFLORA*

Sreepriya T K*, Alan Jacob*, P. Ajith Kumar, Anusree K P, Asiyath Shabana K J, Juzaira T P,
Meenu Aleyas

* Department of Pharmacognosy, Malik Deenar College of Pharmacy Seethangoli, Kasaragod, Kerala

ABSTRACT

Chassalia curviflora is an evergreen shrub grows in tropical region. It is endemic to East Asia. It has potential uses like anti hypertensive, anti bacterial and traditionally used for eye infection, ear infection and insect bites. The present study highlights the phytochemical and invitro pharmacological studies of various extracts of *Chassalia curviflora*. The phytochemical screening of the plant extracts shows the presence of various chemical constituents. The chloroform extract shows potent antioxidant and anti-inflammatory activity when compared to other extracts. The presence of chemical constituents such as alkaloids, phenolics, flavanoids and saponins may be responsible for the anti oxidant and anti-inflammatory activities.

KEYWORDS: *Chassalia curviflora*, antioxidant, anti inflammatory.

Author for correspondence:

Alan Jacob,

Department of Pharmacognosy, Malik Deenar
College of Pharmacy Seethangoli, Kasaragod, Kerala
E mail: alanjacob6@gmail.com

INTRODUCTION

Plants are rich source of variety of chemicals with nutritive and therapeutic properties. Plants are being in medicines from time immemorial because they have fitted the immediate personal need they are accessible and inexpensive. Plant products also be useful as starting material for the semi synthetic preparation of other drugs. Antioxidants are the agents which scavenge free radicals and prevent the damage caused by reactive oxygen species. Antioxidants are absolutely critical for maintaining optimal cellular and systemic health and well being. Anti inflammatory

Agents used to reduce or prevent the inflammation. The signs of inflammation are redness, swelling, heat and pain. The plant *Chassalia curviflora* is endemic to East Asia and it is traditionally used for eye infection, ear infection and insect bites, it also has potential antihypertensive and antibacterial activity.^[1]

MATERIALS AND METHODS

Plant Collection

The leaves of the plant *Chassalia curviflora* [family: Rubiaceae] was collected from Kasaragod and wayanad. The plant material was taxonomically identified by the botanist Mr. Shijith P P, Assistant Professor, Department of botany, Govt. College Kasaragod. The leaves was dried under shade for about 7 days and then powdered with mechanical grinder and stored in an air tight container.

Macroscopic evaluation

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This refers to the organoleptic properties such as color, odour, size, shape and other taxonomical features of the plant. [2,3]

Microscopic evaluation

The transverse section of the leaf and powder of the leaf were used for the study. Both qualitative and quantitative studies were done. [4,5,6]

Phytochemical screening

Various chemical tests were carried out to determine the presence of various chemical constituents such as alkaloids, glycosides, terpenoids, sterols etc. [3,6,8]

Quantitative microscopy

It is carried out to determine the length and width of fibres and stomata present on the leaves of the plant. [4,5,6]

Physico-chemical evaluation

Physico-chemical parameters such as moisture content, ash values (acid insoluble and water soluble ash), extractive values (alcohol and water soluble extractive) and foreign matter were determined. [2,3,6]

In vitro Activity

Invitro antioxidant and anti-inflammatory activity were carried out and determine the IC₅₀ values. [10,11,12,13,14,15,16]

RESULTS AND DISCUSSION

Macroscopic evaluation was done by means of organs of sense. This included evaluation of drug by colour, size, shape and also determine the taxonomical features of the plant. The results are presented in the **table-1** and **figure-1** shows leaf and fruit of the plant *Chassalia curviflora*. The phytochemical screening of the various extracts of plant shows the presence of alkaloids, phenolic compounds, flavanoids, carbohydrate, proteins, terpenoids and saponins which are included in the **table-2**. The physico chemical parameters such as moisture content, ash values, extractive values and foreign matter were determined and the results are shown in the **table-3**. **Figure-2** shows the tranverse section of leaf consists of thick and prominent midrib and thin bifacial lamina. The epidermis of the midrib is fairly thick, squarish in shape with thick walls. The ground tissue of the upper epidermis is parenchymatous. The pallsade layer of lamina is translucent. Ground tissue includes 5 or 6 layers of collenchymatous cell and remaining linear ground tissue is parenchymatous. The vascular system

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consists of wide, deep, bowl shaped vascular strand. The two arms of the vascular strand are incurved forming deep loops. The vascular strand consists of several compact, radial lines of xylem element. All along the lower surface of the xylem strand occurs thin 3 or 4 layers of phloem elements. The leaf powder exhibits fragments of epidermal peelings. The epidermal cells are thick walled, slightly wavy and polygonal in outline. The stomata are paracytic with 1 or 2 pairs of subsidiary cells lying parallel to long axis of the guard cells. The stomata are elliptical. The cells have thick and straight walls. The cells have dense, amorphous mass of protoplasmic content. Calcium oxalate crystals are seen in the ground parenchyma of the midrib and are solitary and diffuse in distribution these are shown in **figure-3**. Quantitative microscopy of leaf were done using projection microscope (ALMICRO-Micro measures and instruments). The eye piece micrometer was calibrated using stage micrometer (calibration factor). The coarsely powdered drug was stained with phlouroglucinol and concentrated hydrochloric acid. Then the length and width of fibres were measured. The results are presented in **table-4**. Invitro antioxidant studies were done by using hydrogen peroxide radical scavenging assay. The extract and standard exhibited dose dependent activity and the IC₅₀ values found out and the results are shown in **table-5**. Invitro anti-inflammatory activity were determined by protein denaturation method and IC₅₀ values were determined and the results are presented in **table-6**.

Table-1 Macroscopic evaluation of *Chassalia curviflora*

Kingdom	Plantae
Phylum	Magnoliophyta
Order	Rubiales
Family	Rubiaceae
Genus	Chassalia
Species	Curviflora
Binomial name	Chassalia curviflora
Plant type	Evergreen shrub
Plant height	1-2 meter
Leaves	Green, opposite, elliptic, obovate, acute (or) acuminate apex
Flower	Pinkish white with yellow

	on a curved floral tube
Fruit	Drupe or ellipsoidal or round, purplish black, two seeded, 5-6 mm wide
Stem	Soft wooded
Calyx	1.5mm long, 5 lobes, ovate
Corolla	8mm long, curved, obtuse
Inflorescence	Cymose



Fig-1 Leaf & fruit of *Chassalia curviflora*

Table-2 Preliminary phytochemical screening of leaves of *Chassalia curviflora*

S l n o	Phytoconstituents	Pet.ether	Chloroform	Ethyl acetate	Methanol	Distilled water
1	Alkaloids	-	+	-	-	+
2	Glycosides	-	-	-	-	-
3	Phenolic compounds	-	+	+	+	+
4	Flavanoids	-	+	+	-	+
5	Carbohydrates	-	-	+	+	+
6	Proteins	-	-	-	+	-
7	Terpenoids	-	-	+	+	+
8	Saponins	-	+	-	+	+



T S of *Chassalia curviflora* leaf

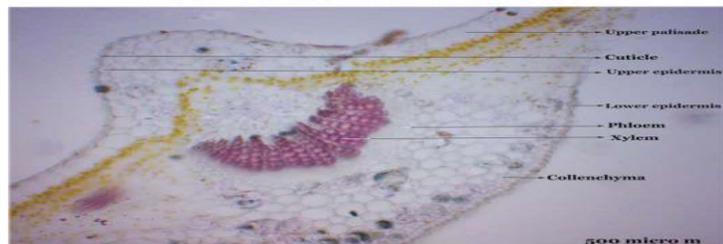


Fig-2 T.S of *Chassalia curviflora*



Tracheids

Stomata



Calcium oxalate crystals

Xylem

Chassalia curviflora leaf powder microscopy

Fig-3 Powder microscopy of *Chassalia curviflora*

Table-3 Physico-chemical evaluation of *Chassalia curviflora*

Sl no	Parameters	Average yield (% W/W)
1	Moisture content	4.56
2	Total ash	13.33
3	Acid insoluble ash	0.81
4	Water soluble ash	7
5	Alcohol soluble extractive value	18.2
6	Water soluble extractive value	27.3
7	Foreign matter	0.363

Table-4 Quantitative microscopy maximum and minimum length and width of fibres of *Chassalia curviflora*

s. no	Parameters of fibres	Average (μm)	Maximum (μm)	Minimum (μm)
1	Length	424.5	707.5	141.5
2	Width	42.45	70.75	14.15

Table-5 Antioxidant activity IC₅₀ values of *Chassalia curviflora*

Sl no	Samples	IC ₅₀ Values
1	Standard	93.35
2	Chloroform	114.7
3	Ethyl acetate	125.7
4	Methanol	128.48
5	Aqueous	137.0
6	Pet. Ether	154.16

Table-6 Antiinflammatory activity IC₅₀ values of *Chassalia curviflora*

Sl no	Samples	IC ₅₀ Values
1	Standard	38.72
2	Chloroform	55.74
3	Ethyl acetate	72.35
4	Methanol	92.5
5	Aqueous	111.08
6	Pet. Ether	145.72

CONCLUSION

The leaves of the plant *Chassalia curviflora* were collected dried and powdered. Macroscopic and microscopic evaluation was performed to identify different macroscopical and microscopical characters. Pharmacognostic studies were carried out and determine the moisture content, Ash value, Extractive value, Percentage of foreign matter. Quantitative microscopical determination of fibres present in the leaves were also carried out. The powdered drug material was subjected to successive solvent

extraction using pet ether, chloroform, ethyl acetate, methanol, and water. The extracts were used for further phytochemical and biological studies. The evaluation of phytochemical studies reveals the presence of phyto constituents like carbohydrate, flavonoids, alkaloids, saponins etc. Further studies were carried out for the determination of invitro antioxidant activity by hydrogen peroxide radical scavenging assay and anti-inflammatory activity by inhibition of protein denaturation method. From both studies chloroform extracts shows maximum activity than other extracts when compared with standard drug, may be due to the presence of flavonoids, saponins and phenolic compounds. As a whole it has been concluded that the leaf of the plant *Chassalia curviflora* on which not much work has been carried out, it has good antioxidant and anti-inflammatory effect. A previous reports also confirms that the above said effects was due to the same compounds present in other plant parts or another plants of the same species. Further studies are required to find out the actual compound responsible for plants medicinal property which can be achieved by isolation of all possible compounds and their pharmacological screening.

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