Contents lists available at ScienceDirect

Asian Pacific Journal of Tropical Biomedicine

journal homepage:www.elsevier.com/locate/apjtb

Document heading

Inter-specific variation studies on the phyto-constituents of *Christella* and *Adiantum* using phytochemical methods

Muraleedharannair Jalajakumari Mithraja[1], Johnson Marimuthu @ Antonisamy[2], Mony Mahesh[1], Zachariah Miller Paul[1], Solomon Jeeva[1]

[1] *Centre for Biodiversity and Biotechnology, Department of Botany,Nesamony Memorial Christian College, Marthandam – 629 165, Tamil Nadu, India*
[2] *Department of Plant Biology and Plant Biotechnology,St. Xavier's College (Autonomous), Palayamkottai, Tamil Nadu, India*

ARTICLE INFO

Article history:
Received 21 December 2011
Received in revised form 29 December 2011
Accepted 15 February 2012
Available online 28 April 2012

Keywords:
Inter- specific variation
Adiantum
Christella
Phytochemistry
Chemical marker

ABSTRACT

Objective: To examine the phyto-constituents of *Adiantum caudataum (A. caudataum), Adiantum latifolium (A. latifolium), Adiantum lunulatum (A. lunulatum), Christella dentata (C. dentate)* and *Christella parasitica (C. parasitica)*, to provide chemical marker and find inter- specific variation between the medicinally important genuses. **Methods:** The dried and powered leaves materials (50 g) were extracted successively with 250 mL of petroleum ether, ethyl acetate, methanol, chloroform, acetone, benzene and water by using Soxhlet extractor for 8 h at a temperature not exceeding the boiling point of the solvent. The aqueous extracts were filtered using Whattman filter paper (No.1) and then concentrated in vacuum at 40 ℃ using Rotary evaporator. The residues obtained were stored in a freezer −70 ℃ until further tests. Phytochemical screening of the extracts was carried out according to the standard methods. **Results:** A total of five plants and 30 extracts were examined for the phytochemical screening. The crude extracts of *A. caudataum, A. latifolium, A. lunulatum, C. dentata* and *C. parasitica* showed varied degree of phyto-constituents with reference to solvents of the plant extracts. A total of three plants and 18 extracts were examined for the phytochemical screening *A. caudataum, A. latifolium, A. lunulatum*. The steroid is totally absent in the tested extracts of A. lunulatum. Similar to that caoboxylic acid also absent in *A. caudatum* and Coumarin showed its presence only in *A. caudatum. A total* of two plants and twelve extracts were examined for the phytochemical properties of Christella. Of which phenol is present in 11 extracts tested. Steroid is present only in one extract of *C. parasitica* and coumarin is present only in *C. parasitica* five extracts. **Conclusions:** The present study result provids a simple and cheap tool (Chemical marker) for the characterization and identification of the medicinally important taxa. With the help the phytochemical screenings we can find inter- specific variation between the medicinally important genuses and easily we can differentiate the *Adiantum* and *Christella* genuses.

1. Introduction

For a long period of time, plants have been a valuable source of natural products for maintaining human health, especially in the last decade, with more intensive studies for natural therapies. About 80% of individuals from developed countries use traditional medicine, which has compounds derived from medicinal plants. Therefore, such plants should be investigated to better understand their properties, safety and efficiency. The use of plant extracts and phytochemicals, both with known antimicrobial properties, can be of great significance in therapeutic treatments. In the last few years, a number of studies have been conducted in different countries to prove such efficiency [1-15]. Many plants have been used because of their antimicrobial properties, which are due to compounds synthesized in the secondary metabolism of plant [16]. Discovery and development of new therapeutic agents is a continuing process. In spite of the fact that, at present, we have at our command a formidable array of modern drugs, the need to discover and invent new agents is genuine and urgent. Adiantum, commonly called as maiden – hair fern, has about 23 species in India widely distributed in the Indian subcontinent. Maximum

*Corresponding author: Department of Plant Biology and Plant Biotechnology, St. Xavier's College (Autonomous), Palayamkottai, Tamil Nadu, India.
Tel: +91 97 86 92 43 34
Fax: + 91 46 22 56 17 65
E-mail: ptcjohnson@gmail.com

number (15 species; 78%) of species occurs on the mountains of south India [17]. All the species are commonly grown as ornamental plants and majority of them are also used in traditional medicines to cure various diseases like cough, fever, skin diseases, catarrhal affection, throat infection, bronchial disorders, dysentery, ulcer, epilepsy, leprosy, biliousness, inflammation, tumors of spleen, liver and other viscera, cold, headache, piles, hair growth etc. It is also considered as tonic and diuretic. *Adiantum lunulatum* Burm. (*A. lunulatum*), Adiantum capillus – veneris L and Adiantum raddianum *C. Presl* are sold in the market by the trade names "Hansraj" and "Paroshan". All the species are collected from the wild and none of the species is under cultivation.

Adiantum caudatum L. (*A. caudatum*), rhizomes (are used for cough and fever. Fronds of *A. caudatum* are used for wound healing. It possesses antihelmintic, antimicrobial activity, antispasmodic, anti-asthmatic etc, *Adiantum latifolium* Lam (*A. latifolium*) possess antinociceptive and anti-inflammatory activities. Ayurvedic Vaidyas describe the plant *Adiantum lunulaturm* Lam (*A. lunulaturm*) as pungent, alexiteric and used for indigestion. The decoction of leaves is useful in dysentery, diseases of the blood, ulcers and erysipeleas. Sporophylls are used in leprosy and erysipelas. The local people in the Aravalli Hills use the decoction of the leaves in cough, asthma and fever. The paste of leaves is used in leprosy and erysipelas. It is also used to overcome hair falling by putting paste of its leave on head for an hour or so before taking bath for a fortnight. In Mt. Abu area, Bheel uses this plant for urinary diseases and nose bleeding. In former the extract of leaves is taken orally and the paste of leaves is applied on the lower portion of stomach, for clear and early release of urine. The leaf extract is put in drops into the nose to stop bleeding, during summer months. A. lunulatum possess antimicrobial property. Leaf and root decoction is used for the chest complaints in Malaya. Fresh leaf decoction is given to cure irregular menstrual cycle. Plant paste is given to women to help them to conceive. Seven triterpenoid are isolated from whole plants of *A. lunulatum*. Christella dentata (Forssk.) Brounsey & Jermy (*C. dentate*) and *Christella parasitica* (L.) H. Lev, (*C. parasitica*) extracts are used to treat gout and rheumatism. The decoction orally administered for 10 days to cure spermatorrhea. Chiristella is possessed antifungal and antibacterial properties [18-27].

A. caudatum L. is largely used as substitute for Adiantum capilllus- veneris L. and there is also possibility for the intentional or unintentional adulteration of common species in the place of rare species. Such adulterant may not have the same medicinal property as in the original species. Moreover species like *A. lunulatum* and *Adiantum raddianum* are polymorphic taxa with many morphotypes, cytotypes and ecotypes [28]. Most of the ferns show positive results with the difference in the degree of bioactivity between species to species. But interestingly, intra-specific difference in biological activity was observed in the case of the fern *C. parasitica* (L.) H. Lev., which is a highly polymorphic species [14]. Some gatherings of this species show more biological activity in contrast to some other gatherings, which are without the same degree of biological activity. In order to understand the reason for such differences in biological activity, morphological, phytochemical and bioactivity studies were carried out among different gatherings based on the information from the literatures. It has been known that the fern *Christella parasitica* (L.) H. Lev. [*Thelypteris parasitica* (L.) Tard.] is a polymorphic species with the variation in size of the entire plant, pubescens, depth of lobing of pinnae etc. In the mean time there are two distinct morphotypes. They are glandular and eglandular morphotypes. In the former, the leaves bear orange coloured, elongated glands on the lower side of the costa, costules and veins, but such glands are absent in the eglandular morphotype. It is also important to note that the glandular morphotype is very rare in occurrence in contrast to the common eglandular morphotype [14]. When the gatherings, with and without biological activities, were subjected to morphological studies, it was confirmed that the glandular morphotype shows remarkable bioactivity [14]. The availability of such a wide range of variants in these species presents a problem in the selection of suitable variant with good medicinal property. Reliable information on the existing genetic variation is required for selection, breeding and conservation programmes of genetic resources. Phytochemical profile is the first hand indication of all the characters. Some ferns belonging to the genera like Adiantum, Blechnum, Christella, Dicranopteris etc., from Tirunelveli Hills of the Western Ghats were screened for their biological activities, particularly for antibacterial activity. But there is no report on the Kanyakumari District, Western Ghats, India. With this knowledge the present study was aimed to examine the phyto-constituents of *A. caudataum*, *A. latifolium*, *A. lunulatum*, *C. dentata* and *C. parasitica*. In addition it will provide chemical marker and inter- specific variation between the medicinally important plants viz., *A. caudataum*, *A. latifolium*, *A. lunulatum*, *C. dentata* and *C. parasitica*.

2. Materials and methods

Healthy, disease free entire plants of *A. caudatum* L, *A. latifolium* Lam and *A. lunulatum* Burm were collected from from Kakachi, Tirunelveli Hills (1 000 m) on the Western Ghats. Voucher specimen has been deposited in Centre for Biodiversity and Biotechnology, Department of Botany, Nesamony Memorial Christian College, Marthandam - 629 165, Tamil Nadu, India. The fresh materials were washed in tap water 5 min and dried using blotting papers. The washed plant materials were air and shade dried for two weeks and pulverized to powder using mortar. The dried and powered

leaves materials (50 g) were extracted successively with 250 mL of petroleum ether, ethyl acetate, methanol, chloroform, acetone, benzene and water by using Soxhlet extractor for 8 h at a temperature not exceeding the boiling point of the solvent. The aqueous extracts were filtered using Whattman filter paper (No.1) and then concentrated in vacuum at 40 ℃ using Rotary evaporator. The residues obtained were stored in a freezer −70 ℃ until further tests [29]. Phytochemical screening of the extracts was carried out according to the standard methods [30-35].

3. Results

In the present study, the phytochemical screening was performed with acetone, benzene, chloroform, ethanol, petroleum ether and aqueous extracts of the whole plants of *A. caudataum*, *A. latifolium*, *A. lunulatum*, *C. dentata* and *C. parasitica*. A total of five plants and 30 extracts were examined for the phytochemical screening. The crude extracts of *A. caudataum* showed varied degree of phyto-constituents with reference to solvents of the plant extracts. The acetone extracts of *A. caudataum* showed maximum presence of phyto-constituents (6/12), next to that chloroform, ethanol and petroleum ether (4/12) followed by aqueous (3/12) (Table 1). The benzene extracts of *A. caudataum* showed only 2 constituents presence. The results of the phytochemical screening revealed that phenol and carbohydrates are present in all the tested extracts of *A. caudataum* except benezene. Tannin also present in all tested extracts of *A. caudataum* except acetone extracts. Saponin is present in chloroform, petroleum ether and benzene extracts of A. caudataum. The acetone extracts of *A. caudataum* showed the carboxylic acid and xanthoprotein presence. Acetone and ethanol extracts of *A. caudataum* showed the coumarin presence. The acetone extracts of *A. caudataum* showed the unique presence of steroids (Table 1).

The crude extracts of *A. latifolium* showed varied chemical profiles with reference to solvents of the plant extracts. The aqueous and chloroform extracts of *A. latifolium* showed maximum occurrence of phyto-constituents (5/12), next to that acetone and petroleum ether (4/12) followed by benzene

Table 1
Phytochemical variation studies on *A. caudatum*, *A. latifolium* and *A. lunulatum* from Western Ghats, Kerala, India.

Compounds	*A. caudatum*						*A. latifolium*						*A. lunulatum*					
	A	B	C	W	E	P	A	B	C	W	E	P	A	B	C	W	E	P
Alkaloids	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Phenols	+++	−	+++	++	++	+++	+++	+	+++	++	++	+++	++	+	+	+	++	+
Flavonoids	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Saponins	−	+++	++	−	−	++	−	+++	+++	−	−	+++	−	++	++	−	−	++
Proteins	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Quinones	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
Steroids	++	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	−
Tannins	−	+	++	+	+	++	+	−	+	++	−	−	−	+	−	+	−	−
Xanthoprotiens	++	−	−	−	−	−	++	−	−	++	−	−	+	−	−	+	−	−
Carboxylic acids	++	−	−	−	−	−	−	−	++	+	−	−	+	−	−	−	++	−
Coumarins	++	−	−	−	+	−	−	−	−	−	−	−	−	−	−	−	−	−
Carbohydrates	++	−	++	++	+	+	+++	−	+++	+++	+++	+	−	−	++	+	++	−
Total	6	2	4	3	4	4	4	2	5	5	2	4	3	3	3	4	3	2

A − Acetone; B − Benzene; C − Chloroform; W − Aqueous; E − Ethanol; P − Petroleum Ether

Table 2
Phytochemical Variation studies on *C. dentata* (Forssk.) Brounsey & Jermy and *C. parasitica* (L.) H. Lev. from Western Ghats, Kerala, India.

Compounds	*C. dentata*						*C. parasitica*					
	A	B	C	W	E	P	A	B	C	W	E	P
Alkaloids	−	−	−	−	−	−	−	−	−	−	−	−
Phenols	+	+++	++	−	++	++	+	+	+	++	+	+
Flavonoids	−	−	−	−	−	−	−	−	−	−	−	−
Saponins	−	+++	++	−	−	++	−	++	+	−	−	+
Proteins	−	−	−	−	−	−	−	−	−	−	−	−
Quinones	−	−	−	−	−	−	−	−	−	−	−	−
Steroids	−	−	−	−	−	−	−	−	−	−	−	+
Tannins	−	+	+	++	−	−	+++	−	−	++	+++	−
Xanthoprotiens	+	−	−	−	−	−	++	−	−	−	−	−
Carboxylic acids	−	−	++	−	−	−	+++	++	−	−	+++	−
Coumarins	−	−	−	−	−	−	−	++	++	+++	++	+++
Carbohydrates	++	+	+++	+	+	−	−	−	+++	++	+	−
Total	3	4	5	2	2	2	4	4	4	4	5	4

A − Acetone; B − Benzene; C − Chloroform; W − Aqueous; E − Ethanol; P − Petroleum Ether

and ethanol (2/12) (Table 1). The phenol is present in all the tested extracts of *A. latifolium*. The steroid is present only in the petroleum ether extracts of *A. latifolium*. The carboxylic acid is present in chloroform and aqueous extracts of *A. latifolium*. The tannin is present in acetone, chloroform and aqueous extracts of *A. latifolium*. The saponin is present in benzene, petroleum ether and chloroform of *A. latifolium*. Xanthopreotein is existed only in acetone and aqueous extracts of *A. latifolium*. Carbohydrate is showed its presence in aqueous, chloroform, petroleum ether, acetone and ethanol extracts of *A. latifolium* (Table 1).

The crude extracts of *A. lunulatum* showed diverse phyto-profiles with reference to solvents of the plant extracts. The aqueous extracts of *A. lunulatum* showed maximum occurrence of phyto-constituents (4/12), next to that acetone, benzene, chloroform and ethanol (3/12) followed by petroleum ether (2/12) (Table 1). The phenol is present in all the tested extracts of *A. lunulatum*. The carboxylic acid is present only in acetone and ethanol extracts of *A. lunulatum*. The tannin is present in benzene and aqueous extracts of *A. lunulatum*. The saponin is present in benzene, petroleum ether and chloroform of *A. lunulatum*. Xanthopreotein is existed only in acetone and aqueous extracts of *A. lunulatum*. Carbohydrate is showed its presence in aqueous, chloroform and ethanol extracts of *A. lunulatum* (Table 1).

The crude extracts of *C. dentata* showed various phyto-constituents with reference to solvents of the plant extracts. The chloroform extracts of *C. dentata* showed maximum occurrence of phyto-constituents (5/12), next to that benzene (4/12) followed by acetone (3/12) (Table - 2). The phenol is present in all the tested extracts of C. dentate except aqueous extract. The carboxylic acid is present only in chloroform extract of *C. dentata*. The tannin is present in chloroform, benzene and aqueous extracts of *C. dentata*. The saponin is present in benzene, petroleum ether and chloroform of *C. dentata*. Xanthopreotein is existed only in acetone extracts of *C. dentata*. Carbohydrate is showed its presence in aqueous, chloroform, benzene, acetone and ethanol extracts of *C. dentata*. (Table 2)

The crude extracts of *C. parasitica* illustrated different phyto-profiles with reference to solvents of the plant extracts. The ethanol extracts of *C. parasitica* showed maximum occurrence of phyto-constituents (5/12), next to that acetone, benzene, chloroform, petroleum ether and ethanol (4/12) (Table 2). The phenol is present in all the tested extracts of *C. parasitica*. The carboxylic acid is present in acetone, benezene and ethanol extracts of *C. parasitica*. The tannin is present in acetone, ethanol and aqueous extracts of *C. parasitica*. The saponin is present in benzene, petroleum ether and chloroform of *C. parasitica*. Xanthopreotein is existed only in acetone extracts of C. parasitica. Carbohydrate is showed its presence in aqueous, chloroform and ethanol extracts of *C. parasitica*. Coumarin is present in ethanol, chloroform, aqueous and petroleum ether extracts of *C. parasitica*. Steroid is present only in petroleum ether extracts of *C. parasitica* (Table 2).

4. Discussion

A total of three plants and 18 extracts were examined for the phytochemical screening *A. caudataum*, *A. latifolium*, *A. lunulatum*. Of which phenol is present in 17 out of 18 extracts tested. Saponin showed its presence in nine extracts, tannin in 10 extracts, carbohydrates in 13 extracts, xanthoprotein in five extracts, carboxylic acid in 4 extracts and steroid and coumarin is present only in two extracts. The phytochemical studies revealed the variation and confirmed the morphological and cytological differences. The steroid is totally absent in the tested extracts of *A. lunulatum*. Similar to that caoboxylic acid also absent in *A. caudatum* and Coumarin showed its presence only in *A. caudatum*.

A total of two plants and twelve extracts were examined for the phytochemical properties of Christella. Of which phenol is present in 11 extracts tested. Saponin showed its presence in 6 extracts, tannin in 6 extracts, carbohydrates in 8 extracts, xanthoprotein in 2 extracts, carboxylic acid in 4 extracts and steroid in one extract of *C. parasitica* and coumarin is present only in *C. parasitica* five extracts. The phytochemical studies revealed the variation and confirmed the morphological and cytological differences. The steroid is totally absent in the tested extracts of *C. dentata*. Similar to that coumarin also absent in *C. dentata* and caroboxylic acid showed its present only in chloroform extracts of *C. dentata* but in *C. parasitica* caroboxylic acid is absent in chloroform extracts and showed its presence in ethanol, acetone and benzene extracts of *C. parasitica*. The present study result provided a simple and cheap tool for the characterization and identification of the medicinally important taxa. With the help the phytochemical screening we can easily differentiate the Adiantum and Christella species.

There are some studies on phytochemistry and pharmacology on *C. parasitica* and *A. lunulatum* [12-14, 18-20], but there is no report on *C. dentata*, *A. caudataum* and *A. latifolium*. In the case of Christella, the present study confirmed the previous observation and provided simple and cheap chemical marker for the differentiation of the medicinally important plants *C. parasitica* from *C. dentata*.

The presence of antimicrobial activity in a particular part of a particular species may be due to the presence of one or more bioactive compounds such as alkaloids, glycosides, flavonoids, steroids, saponins etc. [36]. Recently, a number of plants have been reported for antimicrobial properties across the world [1-16, 18-20]. In the present investigation, five plants from India have been screened for phytochemical properties. The various phytochemical compounds detected are known to have beneficial importance in medicinal sciences. Many tannin-containing drugs are used in medicine as astringent. They are used in the treatment of burns as they precipitate the proteins of exposed tissues to

form a protective covering. They are also medicinally used as healing agents in inflammation, leucorrhoea, gonorrhoea, burns, piles and as antidote. Tannins have been found to have antiviral, antibacterial, antiparasitic effects, anti-inflammatory, antiulcer and antioxidant property for possible therapeutic applications. It was also reported that certain tannins were able to inhibit HIV replication selectively and was also used as diuretic [37,38]. In the present study we revealed the tannins (16/30 extracts) presence in all the tested plants. Saponins are considered a key ingredient in traditional Chinese medicine and are responsible for most of the observed biological effects. Saponins are known to produce inhibitory effect on inflammation. There is tremendous, commercially driven promotion of saponins as dietary supplements and nutriceuticals. Saponin possesses specific physical, chemical and biological activities that make them useful as drugs. Some of these biological properties include anti-microbial, anti-inflammatory, anti-feedent, and hemolytic effects [39]. Saponin is used as mild detergents and in intracellular histochemical staining. It is also used to allow antibody access in intracellular proteins. In medicine, it is used in hypercholestrolaemia, hyperglycaemia, antioxidant, anticancer, antifungal, anti-inflammatory, weight loss, etc. In the present study we observed the saponin (15/30 extracts) also present in all the tested plants. Plant steroids are known to be important for their cardiotonic activities and also possess insecticidal and antimicrobial properties. They are also used in nutrition, herbal medicine and cosmetics. In the present study, steroid (03 / 30) is present all the selected plants except *A. lunulatum* and *C. dentata*. Coumarin has been used as anti-coagulant drugs and to treat lymphedema. In the present study we observed the Coumarin (07/30 extracts) presence in *A. caudatam* and *C. parasitica* Phenolic possesses specific physical, chemical and biological activities that make them useful as drugs. Some of these biological properties include anti-microbial, anti-inflammatory, anti-feedent, anti-viral, anti-cancer, and and vasodilatory actions. In the present study we observed phenolic presence in all tested plant with maximum percentage (28/30). Phytochemical studies on extracts of *A. caudataum*, *A. latifolium*, *A. lunulatum*, *C. dentata* and *C. parasitica* confirmed the presence of carbohydrates, steroids, tannins, saponins, carboxylic acid, coumarins, xanthoprotein and phenolic compounds. The antimicrobial activity of *A. lunulatum* and *C. parasitica* may be due to one/more group of above phyto-constituents. The result obtained in this study suggests a potential application *A. caudataum*, *A. latifolium*, *A. lunulatum*, *C. dentata* and *C. parasitica* for treatment of skin wounds, typhoid, asthma, ulcer, inflammation, weight loss etc. Further benefits include the absence of adverse systemic effects, and a low incidence of resistance. Medicinal plants are of great importance to the health of individual and communities. The medicinal value of these plants lies in some chemical active substances that produce a definite physiological action on the human body.

Conflict of interest statement

We declare that we have no conflict of interest.

References

[1] Singh M, Singh N. Khare PB, Rawat AKS. Antimicrobial activity of some important *Adiantum* species used traditionally in indigenous systems of medicine. *J Ethnopharmacol* 2008; **115** (2): 327-329.

[2] Parihar P, Parihar L, Bohra A. *In vitro* anti-bacterial activity of fronds (leaves) of some important pteridophytes. *J Microbiol and Antimicrobials* 2010; **2**(2): 19-22.

[3] Hassan SW, Umar KA, Dubai YU, Ebbo AA, Faruk UZ. Antibacterial, phytochemical and toxicity studies of *Pteridum aquilinum* L. Dennstaedtiaceae) in rabbits. *J Pharmacol Toxicol* 2007; **2**(2): 168-175.

[4] Pradeep Parihar, Leena Parihar, Achaleshwar Bohra. Antibacterial activity of *Athyrium pectinatum* (Wall.) Presl. *Nat Prod Rad* 2006; **5**(4):262-265.

[5] Haripriya D, Selvan N, Jeyakumar N, Periasamy R, Johnson M, Irudayaraj V. The effect of extracts of *Selaginella involvens* and *Selaginella inaequalifolia* leaves on poultry pathogens. *Asian Pac J Trop Med* 2010; **3**(9): 678-681.

[6] Irudayaraj V, Janaky M, Johnson M, Selvan N. Preliminary phytochemical and antimicrobial studies on a Spike-Moss *Selaginella inaequalifolia* (Hook. & Grev.) Spring. *Asian Pac J Trop Med* 2010; 4957-960.

[7] Singh M, Govindarajan R, Rawat AKS, Khare PB. Antimicrobial flavonoid rutin from *Pteris vittata* L. against pathogenic gastrointestinal microflora. *Am Fern J* 2008; **98**(2):98-103.

[8] Shokeen P, Ray K, Bala M, Tandon V. Preliminary studies on activity of *Ocimum sanctum*, *Drynaria quercifolia*, and *Annona squamosa* against Neisseria gonorrhoeae. *Sex Transm Dis* 2005; **32**(2):106-111.

[9] Khan A, Haque E, Mukhlesur Rahman M, Mosaddik A, Rahman M, Sultana N. Isolation of antibacterial constituent from rhizome of Drynaria quercifolia and its sub-acute toxicological studies. *DARU* 2007; **15**(4): 205-211.

[10] Zakaria ZA, Mat Jais AM, Mastura M, Mat Jusoh SH, Mohamed AM, Mohd Jamil NS, et al. *In vitro* anti-staphylococcal activity of the extracts of several neglected plants in Malaysia. *Int J Pharmacol* 2007;**3**: 428-431.

[11] Maridass M, Ghantikumar S. Antibacterial activity of leaves of *Blechnum orientale* L. *Pharmacologyonline News lett* 2008; **3**:58-60.

[12] Sahayaraj K, Borgio JAF, Raju G. Antifungal activity of three fern extracts on causative agents of groundnut early leaf spot and rust diseases. *J Plant Prot Res* 2009; **49** (2): 141-144.

[13] Manikam VS, Benniamin A, Irudayaraj V. Antibacterial activity of leaf extracts of *Christella paracitica* (L.) Lev. *Indian Fern J* 2005; **4**: 87-88.

[14] Paul Raj K, Irudayaraj V, Johnson M, Patric Raja D. Phytochemical and anti-bacterial activity of epidermal glands extract of *Christella parasitica* (L.) H. Lev. *Asian Pac J Trop Biomed* 2011; **1**(1): 8-11.

[15] NejadBS, Deokule SS. Anti-dermatophytic activity of *Drynaria quercifolia* (L.) J. Smith Jundishapur. J Microbiol 2009; 2(1): 25-30.

[16] Mallikharjuna PB, Rajanna LN, Seetharam YN, Sharanabasappa GK. Phytochemical studies of *Strychnos potatorum* L.f.-A medicinal plant. *E -J Chem* 2007; **4**(4): 510- 518.

[17] Manickam VS, Irudayaraj V. *Pteridophyte flora of the Western Ghats- South India*. New Delhi: BI Publications;1992.

18] Nonato FR, Nogueira TM, De Almeida Barros TA, Lucchese AM, Oliveira CE, Santos RR, et al. Antinociceptive and antiinflammatory activities of *Adiantum latifolium* Lam.: Evidence for a role of IL-1beta inhibition. *J Ethnopharmacol* 2010; Avaliable from : http://www.ncbi.nlm.nih.gov/pubmed/20554010

[19] Niranjan Reddy VL, Ravikanth V, Prabhakar Rao T, Diwan PV, Venkateswarlu Y. A new triterpenoid from the fern *Adiantum lunulatum* and evaluation of antibacterial activity. *Phytochemistry* 2001; **56**(2): 173-175.

[20] Besharat M, Rahimian M, Ghaemi EA, Besharat Sima. Effect of ethanolic extract of Adiantum capillus-veneris in comparison with gentamicine on 3 pathogenic bacteria *in vitro*. *Pharm Sci* 2009; **15**(1):49-52.

[21] Parihar P, Parihar L. Some pteridophytes of medicinal importance from Rajasthan. *Nat Prod Rad* 2006; **5**(4): 297-301.

[22] Benjamin A, Manickam VS. Medicinal pteridophytes from the Western Ghats. *Ind J Trad. Knowl* 2007; **6**(4): 611- 618.

[23] Shil S, Dutta Choudhury M. Ethnomedicinal Importance of pteridophytes used by Reang tribe of Tripura, North East India. *Ethnobotanical Leaflets* 2009; **13**: 634-643.

[24] Rout SD, Panda T, Mishra N. Ethnomedicinal studies on somepteridophytes of Similipal Biosphere Reserve, Orissa, India. *Inte J Med & Med Sci* 2009; **1**(5): 192-197.

[25] Sen A, Ghose PD. A note on ethnobotanical studies of some pteridophytes in Assam. *Ind J Trad Know* 2011; **10**(2): 292-295.

[26] Thulsi Rao K, Reddy KN, Pattanaik C, Reddy CS. EthnomedicinalImportance of Pteridophytes used by Chenchus of Nallamalais, Andhra Pradesh. *India Ethnobotanical Leaflets* 2007; **11**: 6-10.

[27] Maridass M, Raju G. Conservation status of Pteridophytes, Western Ghats, South India. *IJBT* 2010; **1**: 42-57.

[28] Johnson M, Irudayaraj V, Rajkumar SD, Manickam VS. Isozyme Markers for the Crude Drugs of Maiden Hair Ferns from the Western Ghats, South India. *Natural products: An Indian J* 2010; **6**(1):55.

[29] Aiyelaagbe OO, Osamudiamen PM. Phytochemical screening for active compounds in *Mangifera indica*. *Plant Sci Res* 2009; **2**(1): 11-13.

[30] Shyamala Gowri S, Vasantha K. Phytochemical Screening and Antibacterial Activity of *Syzygium cumini* (L.) (Myrtaceae) Leaves Extracts. *IntJ Pharm Tech Res* 2010; **2**(2): 1569-1573.

[31] Aparna Saraf. Phytochemical and Antimicrobial Studies of Medicinal Plant Costus speciosus (Koen.) *E-J Chem* 2010; **7**(S1): S405-S413.

[32] Ngbede J, Yakubu RA, Njam DA. Phytochemical Screening for active compounds in Cornarium schweinfurthii leaves from Jos North, Plateau state. *Nigeria Res J Biol Sci* 2008; **3**(9): 1076-1078.

[33] Onwukeame DN, Ikuegbvweha TB, Asonye CC. Evaluation of phytochemical constituents antibacterial activities and effects of exudates of *Pycanthus angolensis* weld warb on corneal ulcers in rabbit. *Trop J Pharm Res* 2007; **6**(20): 725-730.

[34] Edeogo HO. Phytochemical constituents of some Nigerian medicinal plants. *Afr J Biotechnol* 2005; **4(7)**: 685-688.

[35] Harborne JB. *Phytochemical methods: A guide to modern techniques of plant analysis*. 3rd ed. New York: Chapman and Hall;1998,p. 1-150.

[36] Balandrin MJ, Klocke JA. *Medicinal, aromatic and industrial materials from plants*. Verlag, Berlin, Heidelberg:Bajaj Springer;1988,p. 1-36.

[37] Lü L, Liu SW, Jiang SB, Wu SG. Tannin inhibits HIV-1 entry by targeting gp41. *Acta Pharmacol Sin* 2004; **25** (2): 213-218.

[38] Kolodziej H, Kiderlen AF. Antileishmanial activity and immune modulatory effects of tannins and related compounds on Leishmania parasitised RAW 264.7 cells. *Phytochemistry* 2005; **66** (17): 2056-2071.

[39] George F, Zohar Kerem, Harinder PSM, Klaus Becker. The biological action of saponins in animal systems: a review. *Br J Nutri* 2002; **88** (6): 587-605.