Chapter 3

Review of Literature

The available literature on *C. macrophylla* regarding the traditional use and the plant parts used were reviewed first (Raman Nampoothiri K R, 2006). Published articles regarding phytochemical studies, isolation of the anticancer alkaloid camptothecin, antidiabetic study in *Wistar albino* rats, skeletal muscle relaxant property, anti-amoebic and parasiticidal property were also reviewed.

Available articles on antidiabetic activity, chemical constituents, antibacterial and antifungal activities on *H. arifolia* were reviewed in the next step.

In continuation to the above steps articles available on various anti-diabetic mechanisms of action such as \( \alpha \)-amylase, \( \alpha \)-glucosidase, DPP-IV inhibition, PPAR\( \gamma \) up regulation, Glut-4 translocation, other non-insulin dependent methods of glucose homeostasis, docking and SAR studies, were also reviewed and used for the development of procedures for related experiments.

General methods of isolation, identification and quantification of polyphenolics were reviewed so as to develop methods suitable for the analysis of *C. macrophylla* and *H. arifolia* extracts.

3.1 AVAILABLE DATA ON *C. macrophylla*

1. (Kulkarni AV et al., 2011) There is a mention of the use of *C. grandiflora* in the Ayurvedic preparation ‘Kumaryasavam’ (gynaecological disorders) and ‘Sudarsanasavam’ (memory enhancers). Entire plant and roots are used for antipyretic effect and stomach disorders. The plant is useful in the treatment of skin diseases and inflammations. The percentage of total alkaloids was found to be more in the stem with bark as compared to the leaves. Ethanol extracts of stem with bark and callus were tested for their antimicrobial activity. Callus extracts containing alkaloids exhibited good antimicrobial activity against gram positive organisms comparable with that of cephotoxime, the standard antibiotic.
2. (Kulkarni A V et al., 2010) Phytochemical investigations were revealed the presence of steroidal alkaloids, such as chonemorphine and funtumafine in *C. grandiflora*. Campothecin, a well-known anticancer alkaloid were detected in ethanol extracts of stem with bark and cultures derived from *C. grandiflora*. Campothecin content was 0.013 mg/g in stem with bark and 0.003 mg/g in callus.

3. (Shende V S et al, 2009) In the light of the traditional claim of the antidiabetic property of *C. fragrans*, the study was performed on *Wistar albino* rats using alcoholic extract of the roots. Diabetes was induced in rats with alloxan and the extract was given per oral (dose-100 mg and 200 mg/kg body weight) during the twelve day treatment regimen. Normal and diabetic rats were treated at the same period. The alcoholic extract at 200 mg/kg dose was found to cause a decrease of the elevated blood sugar in the diabetic animals.


5. (Ranendra Kumar Roy et al., 2005) The skeletal muscle relaxant effect of alcoholic extract of *C. macrophylla* was studied on isolated frog rectus abdominus muscle, isolated rat phrenic nerve diaphragm muscle preparation and in intact young chicks. The parameter studied in the isolated muscle or isolated nerve muscle preparations was the extent of inhibition of acetylcholine or electrically-induced contraction of skeletal muscles. In all the experiments, the effect of drug was compared with that of gallamine and succinylcholine. The alcoholic extract of *C. macrophylla* reduced the acetylcholineinduced contraction of isolated frog rectus abdominus and electrically stimulated contractions of rat phrenic nerve diaphragm in a dose-dependent manner. In unanaesthetized chicks, it produced spastic type of paralysis with extension of neck and limbs. The effects were similar to that of succinylcholine but different from those of gallamine.
6. (Chatterjee D K etal., 1987) Chonemorphine, a steroidal alkaloid isolated from *C.fragrans*(Apocyanaceae) was identified as an anti-amoebic principle during the course of a screening programme for novel anti-parasitic agents from plant sources. At a dosage of 100 mg/kg (x4) chonemorphine led to a 100% cure of experimental hepatic infection in golden hamsters and cleared 90% of intestinal infection in weaning *Wistar* rats at 200 mg/kg (x4) dosages. The discovery of chonemorphine as an anti-amoebic agent is an addition to the few known plant amoebicides such as emetine and conessine.

7. (Shrivastava etal., 1984) Chemical examination of the fat from the root of *C. macrophylla*. Root bark lipids of *C. macrophylla* were investigated. The root contained 0.54% lipids, as glycerides of lauric acid (11.9), stearic acid (13.3), arachidic acid (17), myristic acid (20.2), palmitic acid (10.4), oleic acid (9.6), linoleic acid (11.8) and ricinoleic acid (5.2). Found also were octacosanol, ceryl alcohol, β-Sitosterol and taraxasterol.

3.2 WORKS REPORTED ON *H. arifolia*

1. (Kumudhavalli M V, Jayakar B, 2012) *H. arifolia*, a folklore antidiabetes fern, was evaluated for its antidiabetic properties using rats. Glucose lowering effect and antidiabetes activity were studied in normal rats and streptozotocin (60 mg/kg) induced diabetic rats, respectively. 48 hours after STZ induction, diabetic rats received Ethanolic and aqueous extracts of *H. arifolia* orally at 250 mg/kg and 500 mg/kg body weight daily for 15 days. Glibenclamide (5mg/kg p.o) was used as reference drug. Blood glucose levels were measured on 0th, 2nd, 5th, 10th and 15th days of the study. Ethanolic and aqueous extracts were found to lower the levels of blood glucose in glucose fed rats. All the extracts were evaluated for preliminary phytochemical screening. When the ethanolic and aqueous extracts showed the presence of flavonoids, carbohydrates, phenolic compounds and sterols were the major phyto constituents present in the above fractions. This fraction containing flavonoids is believed to have showed the antidiabetic activity in streptozotocin induced diabetic rats as judged from blood glucose levels, body weight, biochemical parameters and histopathological studies.
2. (Hima Bindhu S V S L N et al., 2012) *H. arifolia* (Burm) Moore of family *Hemionitidaceae* is one of the endemic and widely distributed species on Tirumala hills of Tirupathi, Andhra Pradesh, India. Ethno-medicinally, the genus is important and popularly known as *Ramabanum*. It has been used in burns, menstrual disorders, antiflatulence and antifertility. Eight different organic solvents were used to extract the bioactive compounds from the whole plant to screen the phytochemical and antibacterial activity against infectious disease causing bacterial pathogens such as *Enterobacter aerogenes*, *Klebsiella pneumonia*, *Salmonella paratyphi* A, *Ralstonia eutropha*, *Salmonella typhi*, *Salmonella paratyphi* B, *Staphylococcus aureus*, *Bacillus cereus*, *Bacillus subtilis*, *Bacillus sphericus*, *Bacillus steroothermophilus* and *Micrococcus luteus* by agar well diffusion method. Phytochemical screening showed the presence of flavonoid, steroids and glycosides along with reducing sugar in all the extracts investigated. Gram-negative bacteria such as *Salmonella typhi*, *S. paratyphi* A and *Enterobacter aerogenes* were more susceptible to the crude extracts than gram-positive bacteria. Hence, at any rate *H. arifolia* is an attractive material for further research leading to possible drug development.

3. (Joydip Karmakar and Subhara Kanti Mukhopadhaya, 2011). Antibacterial and antifungal activity of the crude extract, alcoholic extract and extracted phenol from various parts of tropical pteridophyte, *H. arifolia* were tested by agar diffusion and tube dilution assay. Both the crude and alcohol extracts of vegetative and reproductive leaves of *H. arifolia* showed considerable antibacterial activity against gram negative test strain of *Escherichia coli* (MTCC-739). Extract from reproductive leaves also showed moderate antibacterial activity against *Bacillus subtilis* (MTCC-441) (gram positive test strain) but didn’t show any antifungal activity against *Candida albicans* (MTCC-7535).

4. (Anburaja V et al., 2011) The ethanol extract of the pteridophyte *H. arifolia* at 25, 50 and 100 µl were screened for antimicrobial activity by using Kirby Bauer agar well diffusion assay method against the bacterial and fungal strains. Maximum inhibitory (Muraleedharan Nair Jalajakumari Mithraja
etal.,2012) zone was observed at 100 µl against *Salmonella* with the inhibition zone 2.16 mm and no inhibitory action was observed against *Salmonella*, *Candida* and *Trycophyton* at 25 µl.

5. (Shayaraj K et al., 2009) Antimycotic activity of three fern extracts from *H. arifolia*, *Pteridium aquilinum* and *Christella parasitica* were evaluated against groundnut early leaf spot and rust disease causative agents viz. *Puccinia arachidis* Speg. and *Phaeoisariopsis personata* Vanarx, respectively by mycelia dry weight method. Both the fungi were sensitive to all the three fern extracts tested. Among the extracts, chloroform extract of *H. arifolia* was found to have maximum antifungal activity against both fungi. Between these two fungi, *P. personata* was found to be more sensitive to the tested plant extracts than *P. arachidis*. From the study it is concluded that chloroform extract from *H. arifolia* can be utilized for managing rust and leaf spot diseases on groundnut.

6. (Ajikumaran NairS. et al., 2006) *H. arifolia*, a folklore antidiabetes fern, was evaluated for its hypoglycaemic and anti-diabetic properties using rats. Glucose lowering effect and antidiabetes activity were evaluated using glucose tolerance test in normal rats and alloxan diabetic rats, respectively. When different extracts were tested, the ethanol and to some extent, the water extracts were found to lower the levels of blood glucose in glucose fed rats. The ethanol extract showed optimum activity at 200 mg/kg. The extract exhibited only marginal hypoglycaemic activity in overnight fasted normal rats and it was devoid of conspicuous toxic symptoms in sub-acute toxicity evaluation in mice.

7. (David E Giannasi and John T Mickel, 1979) Systematic implications of flavonoid pigments in the fern genus *Hemionitis* (Adiantaceae). The flavonoid chemistry of these taxa coincides with spore type, i.e., taxa from both genera which possess crested spores produce kaempferol and quercetin-3-O-glycosides, while species with tuberculate spores produce only quercetin-3, 4’-O-glycosides.