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# STUDIES ON PHYTOCHEMICAL ANALYSIS OF METHANOLIC EXTRACT OF *CTENOLEPIS GARCINII* (BURM. F.) C.B. CLARKE

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**ABSTRACT**

The phytochemical analysis of methanolic extract of *Ctenolepis garcinii* (Burm. f.) C.B. Clarke was studied in the present study. The methanolic extract was screened using UV-Vis spectroscopy, HPLC and FTIR by standard procedure. The UV-Visible spectrum showed the compounds separated at the nm of 200, 250, 331, 398, 433, 450, 500, 550, 606, 659, 700, 750, 862, 909, 975, 996, 1053 and 1100 with the absorption 4.000, 0.384, 1.342, 1.207, 4.000, 4,000, 4,000, 2.960, 3.612, 0.424, 0.954, 0.331, 0.130, 0.127, 0.111, 0.104, 0.043 and 0.003 respectively. The qualitative HPLC fingerprint profile displayed eight compounds at different retention times. The profile displayed four compounds at different retention times of 1.867min, 2.063min, 2.797min and 3.000min. The profile displayed six prominent peaks at the retention time of 1.963min, 2.107min, 2.283min, 2.647min, 2.793min, 3.000min, followed by two moderate peaks were also observed at the retention time of 3.240min and 3.413min. The result of FTIR analysis was found the presence of functional groups such as ethers, alcohols, solfonic acids, solfonic acids, aliphatic ethers, vinyl ethers, isopropyl group, benzophenones, aldehydes, phosphines, aliphatic compounds, carboxylic acids, aromatic amines, primary amines and amides.

**Key words:** *Ctenolepis garcinii*, Methanolic extract, UV-Visible, HPLC, FTIR.

## INTRODUCTION

For many years, plant source are used by human to alleviate or cure illnesses. Plants constitute a source of novel chemical compounds which are of potential use in medicine and other applications. Plants are occupying important position in allopathic medicine, herbal medicine, homoeopathy and aromatherapy [1]. Medicinal plants are the sources of many important drugs of the modern world. Many of the indigenous medicinal plants are used as spices and food plants; they are also sometimes added to foods meant for pregnant mothers for medicinal purpose [2].

Many plants are cheaper and more accessible to most people especially in the developing countries than orthodox medicine and there is lower incidence of adverse effects after use. These reasons might account for their worldwide attention and use [3]. Medicinal plants have been of age long remedies for human diseases because they contain components of therapeutic value [4]. Some of them are also used for prophylactic purposes. An increasing interest in herbal remedies has been observed in several parts of the world and many of the herbal remedies have been incorporated into orthodox medicinal plant practice. In this background, the present investigation was studied to screen the phytochemicals present in methanolic extract of *Ctenolepis garcinii* (Burm. f.) C.B. Clarke.

## MATERIALS AND METHODS

### Collection of plant materials

The selected plant material in the present study is *Ctenolepis garcinii* (Burm. f.) C.B. Clarke (Figure 1) belonging to the family Cucurbitaceae which was collected from near to Manonmaniam Sundaranar University, Tirunelveli, located in Tirunelveli district, Tamil Nadu, India during the month of December, 2016, identified and confirmed by the flora of the Presidency of Madras (Gamble, 1919). The collected materials were washed thoroughly with tap water to remove the sediment particles. Followed by, the samples were brought in polythene bag to the laboratory, followed by washed using

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distilled water. They were stored in refrigerator for further use.

**Preparation of extracts**

For the preparation of methanolic extract, the plant specimens were washed thoroughly and placed on blotting paper and spread out at room temperature in the shade condition for drying. The shade dried samples were grounded to fine powder using a tissue blender. The powdered samples were then stored in the refrigerator for further use. 30g powdered samples were packed in Soxhlet apparatus and extracted with methanol for 8h separately [5].

**UV-Vis spectral analysis**

The methanol crude extract containing the bioactive compound was analyzed UV-Vis spectroscopically for further confirmation. The methanol crude extract of *Ctenolepis garcinii* (Burm. f.) C.B. Clarke was scanned in a wavelength ranging from 200-1100nm using a Shimazdu spectrophotometer and characteristic peaks were detected [6].

**HPLC Analysis**

The HPLC method was performed on a Shimadzu LC-10AT VP HPLC system, equipped with a model LC-10AT pump, UV-Vis detector SPD-10AT, a Rheodyne injector fitted with a 20µl loop and an auto injector SIL-10AT. A Hypersil® BDS C-18 column (4.6 × 250mm, 5µm size) with a C-18 guard column was used. The elution was carried out with gradient solvent systems with a flow rate of 1ml/min at ambient temperature (25-28°C). The mobile phase consisted of 0.1% v/v methanol (solvent A) and water (solvent B). The mobile phase was prepared daily, filtered through a 0.45µm and sonicated before use. Total running time was 15min. The sample injection volume was 20µl while the wavelength of the UV-Vis detector was set at 254nm [7].

**Instrumentation**

An isocratic HPLC (Shimadzu HPLC Class VP series) with two LC- 0 AT VP pumps (Shimadzu), a variable wave length programmable photo diode array detector SPD-M10A VP (Shimadzu), a CTO- 10AS VP column oven (Shimadzu), a SCL-10A VP system controller (Shimadzu), a reverse phase Luna 5µl C18 (2) and Phenomenex column (250 mm X 4.6mm) were used. The mobile phase components methanol: water (45:55) were filtered through a 0.2µ membrane filter before use and were pumped from the solvent reservoir at a flow rate of 1ml/min which yielded column backup pressure of 260-270kgf/cm$^2$. The column temperature was maintained at 27°C. 20µl of the respective sample and was injected by using a Rheodyne syringe (Model 7202, Hamilton).

**FTIR analysis**

The methanolic extract of *Ctenolepis garcinii* (Burm. f.) C.B. Clarke was shade dried and FTIR analysis was performed using Perkin Elmer Spectrophotometer system, which was used to detect the characteristic peaks and their functional groups. The peak values of the FTIR were recorded. Each and every analysis was repeated twice and confirmed the spectrum [8].

**RESULTS AND DISCUSSION**

**UV-Visible spectrum analysis**

The UV-Visible spectrum of the methanol extract of *Ctenolepis garcinii* (Burm.f) C.B. Clarke was selected at the wavelength of 200nm to 1100nm due to the sharpness of the peaks and proper baseline. The profile showed the compounds separated at the nm of 200, 250, 331, 398, 433, 450, 500, 550, 606, 659, 700, 750, 862, 909, 975, 996, 1053 and 1100 with the absorption 4.000, 0.384, 1.342, 1.207, 4.000, 4,000, 4,000, 2.960, 3.612, 0.424, 0.954, 0.331, 0.130, 0.127, 0.111, 0.104, 0.043 and 0.003 respectively (Figure 2).

**HPLC analysis**

The qualitative HPLC fingerprint profile of the methanolic extract of *Ctenolepis garcinii* (Burm.f) C.B. Clarke was selected at a wavelength of 660nm due to the sharpness of the peaks and proper baseline. The methanol extract prepared by hot extraction was subjected to HPLC for the separation and identification of constituents present in the *Ctenolepis garcinii* (Burm.f) C.B. Clarke. Eight compounds were separated at different retention time of 1.963min, 2.107min, 2.283min, 2.647min, 2.793min, 3.000min, 3.240min and 3.413min respectively. The profile displayed six prominent peaks at the retention time of 1.963min, 2.107min, 2.283min, 2.647min, 2.793min and 3.000min, followed by two moderate peaks were also observed at the retention time of 3.240min and 3.413min (Figure 3).

**FTIR ANALYSIS**

The FTIR spectrum was used to identify the functional group of the active components based on the peak value in the region of infra red radiation. The crude methanol extract of *Ctenolepis garcinii* (Burm.f) C.B. Clarke was passed into the FTIR and the functional groups of the components were separated based on its peak ratio. As illustrated in Table 1 and Figure 4, FTIR spectrum of methanol extract showed different peaks at 529.42, 676, 1039.59, 1073.31, 1163.96, 1214.11, 1383.83, 1643.24, 1737.74, 2359.74, 2851.56, 2920.99 and 3381.95cm$^{-1}$. It was confirmed the presence of functional groups such as ethers (C-O-C bend), alcohols (C-O-H bending), solfonic acids ($SO_3$ sym stretch), solfonic acids ($SO_3$ sym stretch), aliphatic ethers (C-O-C antisym stretch), vinyl ethers (C-O-C antisym stretch), isopropyl group ($CH_3$ deformation), benzophenones (C=O stretch), aldehydes (C=O stretch),

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phosphines (P-H stretch), aliphatic compounds (CH antisym), carboxylic acids (H-bonded OH stretch) and aromatic amines, primary amines and amides ($NH_2$ stretch).

**Figure 1. Natural habit of *Ctenolepis garcinii* (Burm. f.) C.B. Clarke**

**Figure 2. UV-Visible spectrum of methanolic extract of *Ctenolepis garcinii* (Burm.f) C.B. Clarke**

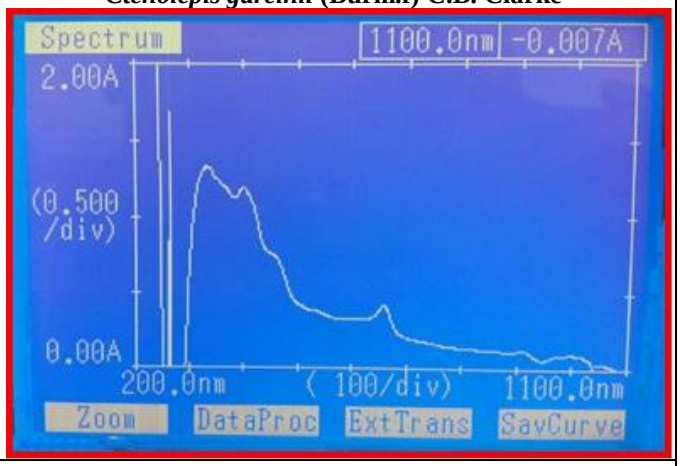



**Figure 3. HPLC analysis of methanol extract of *Ctenolepis garcinii* (Burm.f) C.B. Clarke**

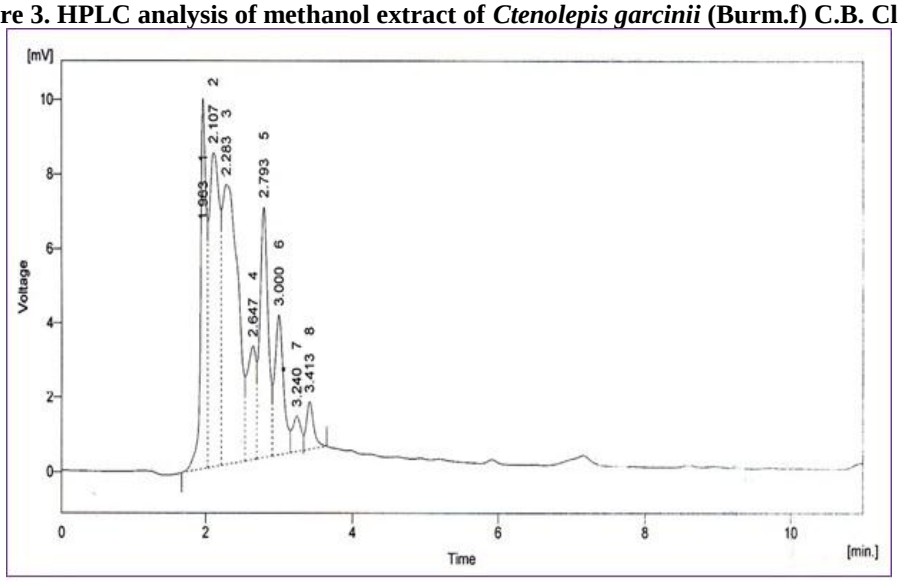
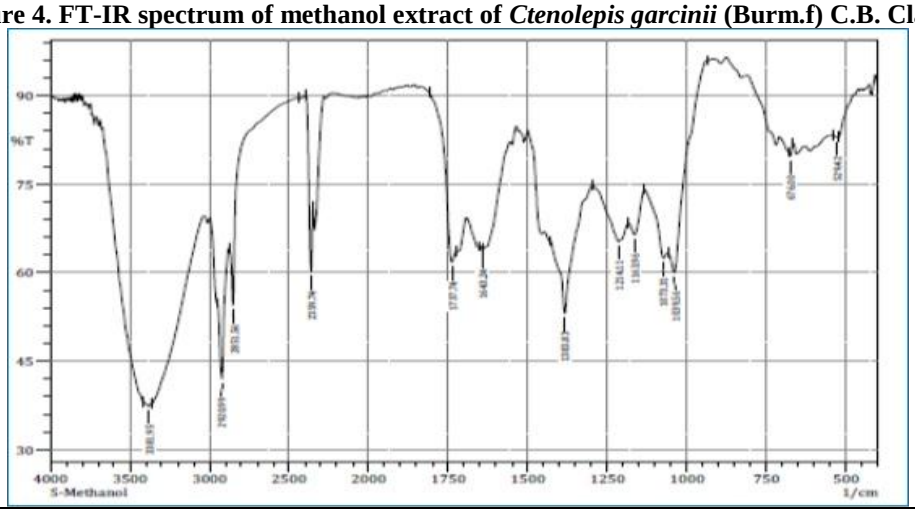



**Figure 4. FT-IR spectrum of methanol extract of *Ctenolepis garcinii* (Burm.f) C.B. Clarke**



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## CONCLUSION

From the present study, it was concluded that UV-Visible spectrum showed the compounds separated at the nm of 200, 250, 331, 398, 433, 450, 500, 550, 606, 659, 700, 750, 862, 909, 975, 996, 1053 and 1100 with the absorption 4.000, 0.384, 1.342, 1.207, 4.000, 4,000, 4,000, 2.960, 3.612, 0.424, 0.954, 0.331, 0.130, 0.127, 0.111, 0.104, 0.043 and 0.003 respectively. The qualitative HPLC fingerprint profile displayed eight compounds at different retention times. The profile displayed four compounds at different retention times of 1.867min, 2.063min, 2.797min and 3.000min. The profile displayed six prominent peaks at the retention time of 1.963min, 2.107min, 2.283min, 2.647min, 2.793min and 3.000min, followed by two moderate peaks were also observed at the retention time of 3.240min and 3.413min. The result of FTIR analysis was found the presence of functional groups such as ethers, alcohols, solfonic acids, solfonic acids, aliphatic ethers, vinyl ethers, isopropyl group, benzophenones, aldehydes, phosphines, aliphatic compounds, carboxylic acids, aromatic amines, primary amines and amides.

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