

ISOLATION AND CHARACTERIZATION OF CAFFEIC ACID FROM THYME SEEDS

**RESEARCH ARTICLE**

**Dr. V. M. MOUNNISSAMY\*<sup>1</sup>, B. PRIYA<sup>2</sup>**

<sup>1</sup>Associate Professor, Department of Pharmaceutical Chemistry,  
College of Pharmacy, Mother Theresa Post Graduate and Research Institute of Health Sciences,  
Gorimedu, Puducherry, India.

<sup>2</sup>Department of Pharmaceutical Chemistry,  
College of Pharmacy, Mother Theresa Post Graduate and Research Institute of Health Sciences,  
Gorimedu, Puducherry, India.

(Received on: 20-04-2020; Revised & Accepted on: 06-07-2020)

---

**ABSTRACT**

Caffeic acid is a dihydroxycinnamic acid belonging to the phenolic acid family, which has a phenyl propanoid structure. It occurs naturally in the wide range of food items such as vegetables, fruits etc. Thyme species are widely used for medicinal purposes. Thyme seeds contain many flavonoids and several phenolic compounds. The present research article discusses about isolation of caffeic acid from thyme seeds and to elucidate its structure using UV, IR<sup>13</sup>, C-NMR, <sup>1</sup>H-NMR AND MASS SPECTROSCOPY.

**Key words:** caffeic acid, thyme, flavonoids, isolation, elucidation, spectroscopy

---

**INTRODUCTION**

In the recent years, there is an increased demand for the food which contains ingredients that may provide health benefits, besides nutrition. (Nanasombat *et al.*). Thyme essential oil constitutes raw material in perfumery and cosmetics due to their aroma flavor (Grigore *et al.*).

Thyme contains many flavonoids, phenolic anti-oxidants like zeaxanthin, lutein, pigenin, naringenin, luteolin and thymonin. Fresh thyme is packed with minerals and vitamins that are essential for optimum health. Thyme provides 0.35 mg of vitamin B-6 or pyridoxine furnishing about 27% of daily recommended intake. (Sharangi AB *et al.*)

Consumption of natural foods rich in flavonoids helps protect from lung and oral cavity cancer. (Aksel B)

Thyme has been used in traditional medicine for the treatment of several respiratory diseases like asthma and bronchitis (Ocana A and Reglero *et al.*). In addition to this, it has several properties, including, antiseptic, antispasmodic, anti-tussive, anti-oxidative, anti-viral (Prashanth R *et al.*) and anti-microbial (Boruga *et al.*).

The present study aimed to isolate the polyphenol caffeic acid from thyme seeds and to elucidate its structure using various spectrometric methods.

**Experimental procedure**

**Extraction**

About 500 grams of the thyme seeds were soaked in a round bottom flask by using hydro-alcoholic solution (70:30) for about 24 hours. The content of the round bottom flask was extracted by reflux for 2 hours. The extract was filtered off and filtrate was collected separately. The filtrate was treated with equal quantity of ether, ethyl acetate and n-butanol and each extract was collected and concentrated separately.

---

**Corresponding Author: Dr. V. M. Mounnissamy\*<sup>1</sup>,**

<sup>1</sup>Associate Professor, Department of Pharmaceutical Chemistry,  
College of Pharmacy, Mother Theresa Post Graduate and Research Institute of Health Sciences,  
Gorimedu, Puducherry, India.

### Purification by column chromatography

Mobile phase as chloroform: ethyl acetate in the ratio of 100, 90:10, 80:20, 70:30, 60:40, 50:50 were taken and Stationary phase as silicagel G. The concentrated organic layers were allowed to run through the column and different fractions were collected and are tested for the presence of flavonoids. The fraction 70:30 and 60:40 showed the positive test for the presence of flavonoids. These two layers were concentrated by using distillation (solvent recovery) and evaporated to get the crude caffeic acid.

### Characterization of isolated compound

#### Caffeic acid

Crystallized from MeOH as pale yellow needles, C<sub>9</sub>H<sub>8</sub>O<sub>4</sub>, Melting point 210-212°C, gave brisk effervescence with saturated NaHCO<sub>3</sub> solution, light blue with Fe<sup>3+</sup> and decolourised Br<sub>2</sub> water.

It was blue under UV and deep blue under UV/NH<sub>3</sub>. Shows positive for shinoda test.

**IR analysis** ( $\gamma_{\max}$ , cm<sup>-1</sup>, KBr) (Figure-2.) 3427, 2558, 1654, 1605, 1534, 1449, 1283, 1217, 1176, 1110, 969, 901, 851, 809.

**<sup>13</sup>C NMR** (500MHz, DMSO-d<sub>6</sub>,  $\delta$ , ppm) (Figure-3) 168.51(s,>C=O); 148.64 (s, C- $\beta$ ); 146.05 (C-3); 145.16 (C-4); 126.19 (C-1); 121.79 (C-6); 116.25 (C-5); 115.59 (C-2); 115.0 (C- $\alpha$ ).

**<sup>1</sup>H NMR** (500MHz, DMSO-d<sub>6</sub>,  $\delta$ , ppm) (Figure- 4) 7.38 (d, J=16.0 Hz, 1H, H- $\alpha$ ), 6.99 (d, J=2.3 Hz, 1H, H-2), 6.92 (dd, J=8.4 & 2.3 Hz, 1H, H-6); 6.72 (d, J=8.4 Hz, 1H, H-5); 6.15 (d, J=16.05 Hz, 1H, H- $\beta$ )

**MASS SPECTRA** (Figure: 5) 180 (M<sup>+</sup>, 100); 181 (M+H); 179 (M-H)

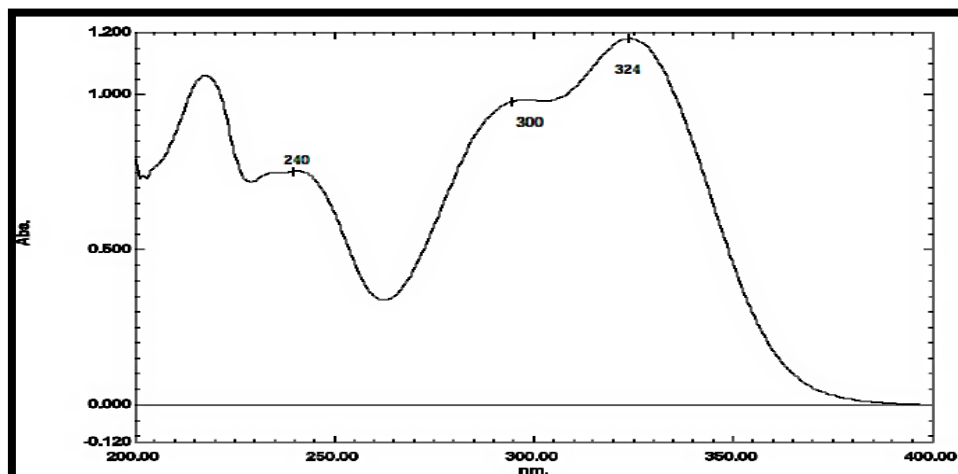


Figure-1: UV spectrum of the compound

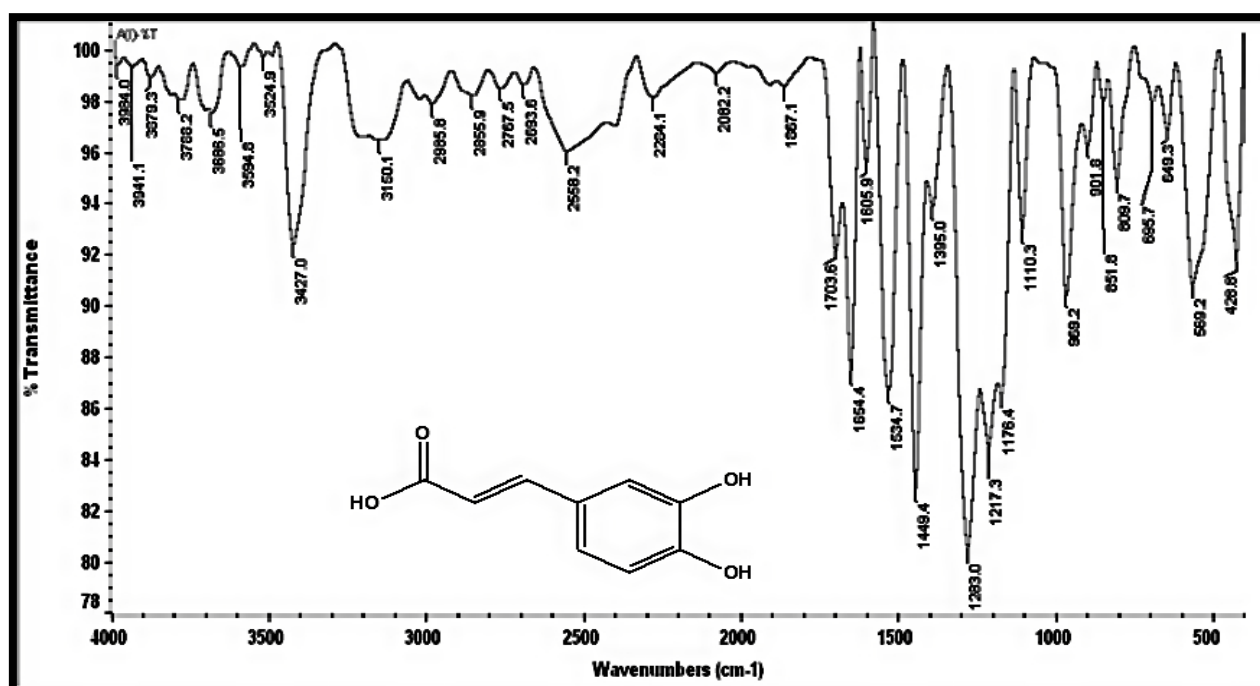


Figure-2: IR spectrum of the compound

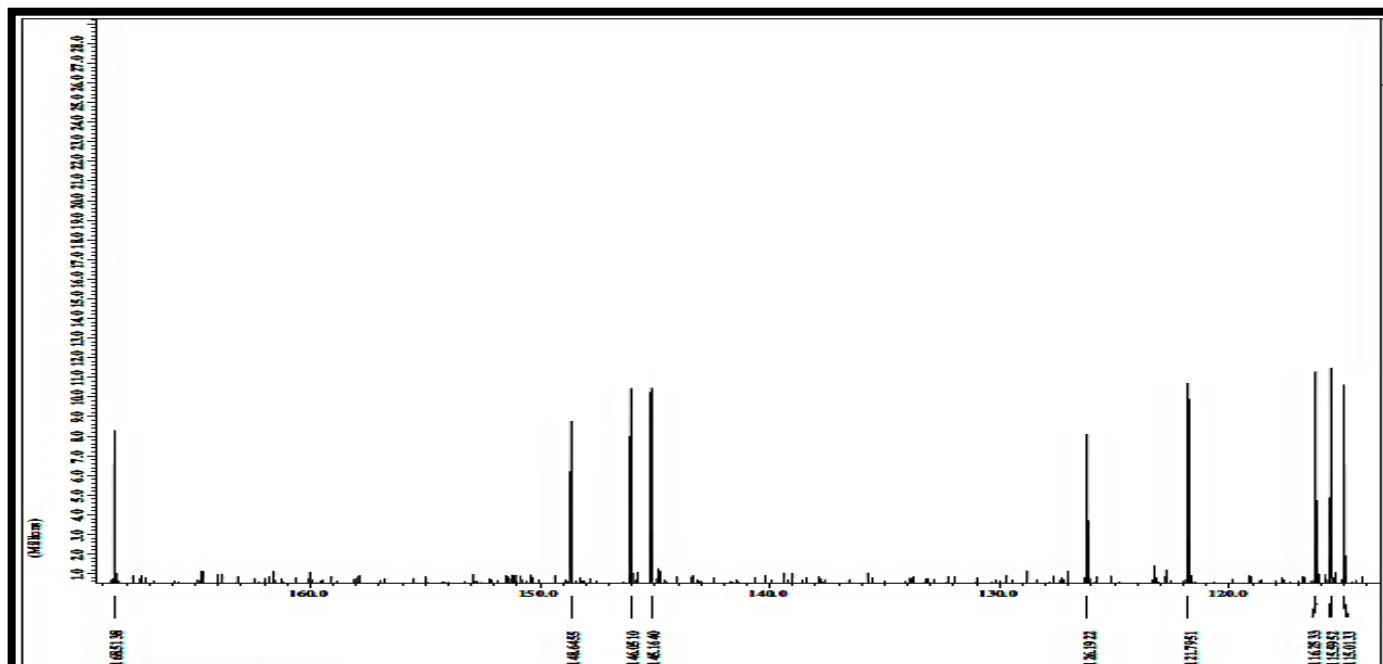


Figure-3: <sup>13</sup>C NMR spectrum of the compound

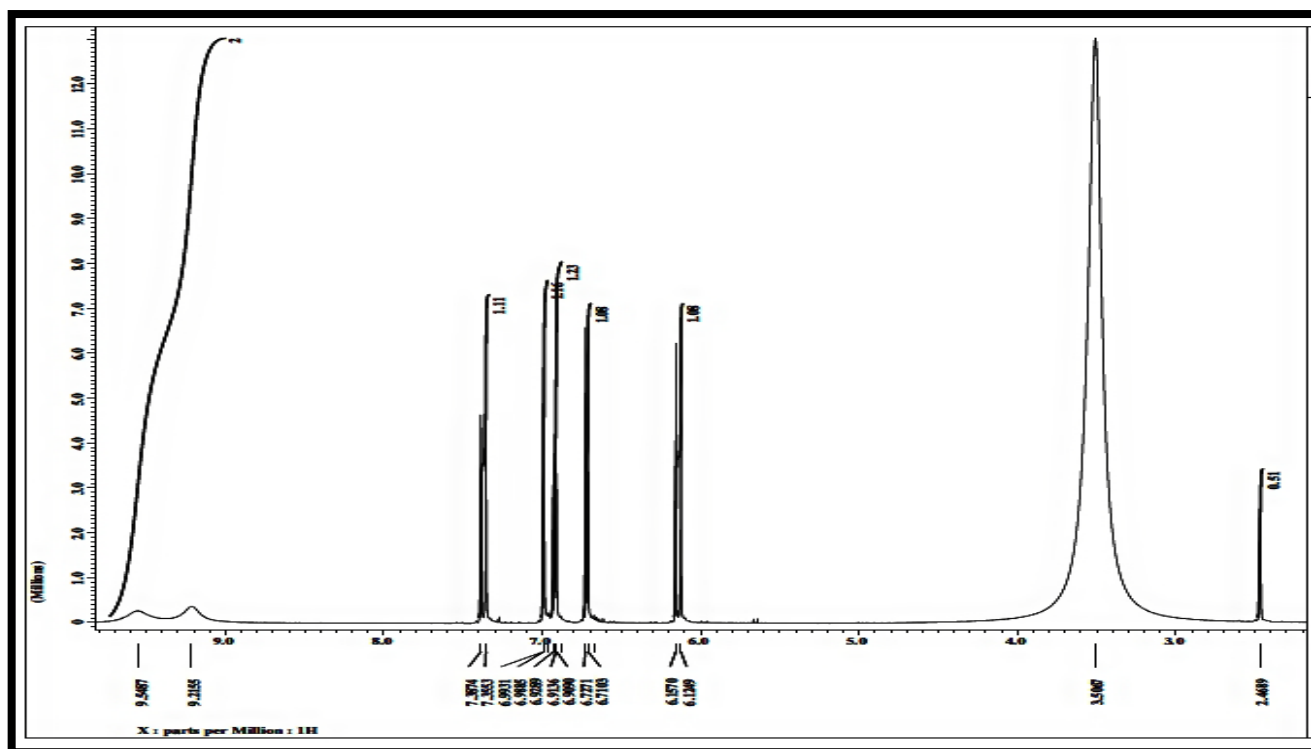


Figure-4: <sup>1</sup>H NMR spectrum of the compound

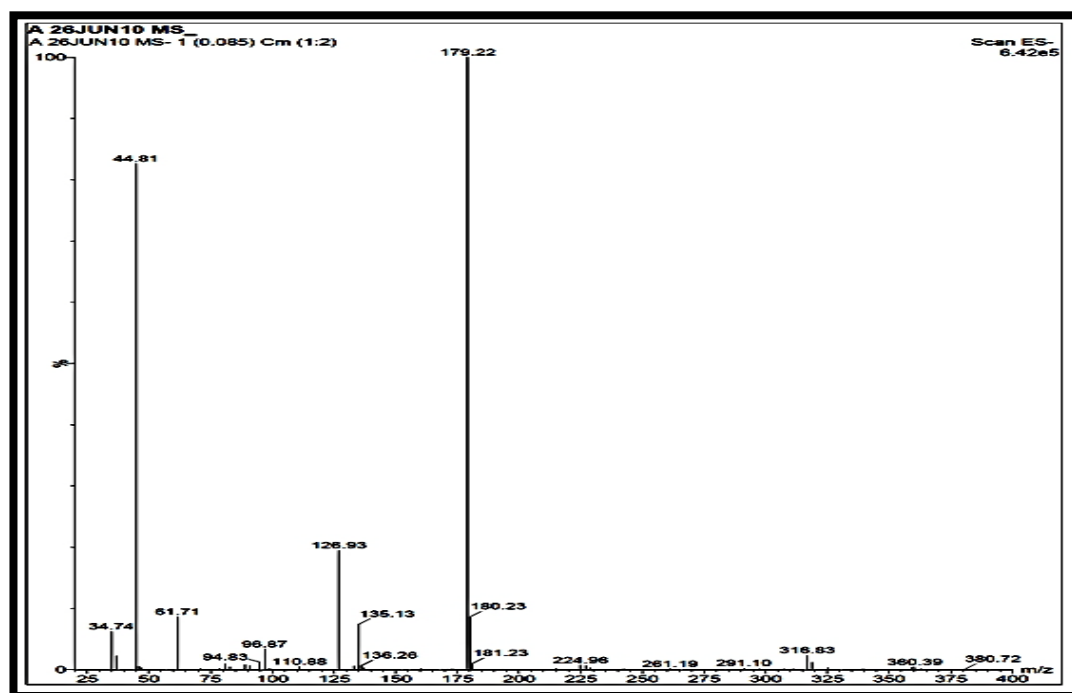


Figure -5: Mass spectrum of the compound

## RESULTS AND DISCUSSION

The crude caffeic acid is isolated from the thyme seeds and its yield was about 0.779 grams. The structural identification of the isolated compound was carried out by MASS SPECTROMETER, <sup>13</sup>C-NMR, <sup>1</sup>H-NMR, IR and UV SPECTROMETER. From the above given spectral data, it is identified as a caffeic acid.

## ACKNOWLEDGEMENT

I Extend my thanks to The Dean, Mother Theresa Post Graduate and Research Institute of Health Sciences, The Guide, Professor and Head, Department of Pharmaceutical chemistry.

## REFERENCES

1. Aksel B. Bioactive compounds in plants – benefits and risks for man and animals. The Norwegian Academy of Science and Letters, Oslo 2008, 13-14.
2. Boruga O, Jianu C, Mișcă C, Goleț I, Gruia AT, Horhat FG. Thymus vulgaris essential oil: chemical composition and antimicrobial activity. J Med Life. 2014; 7( 3): 56-60
3. Grigore A, Paraschiv INA, Colceru-mihul S, Bubueanu C, Draghici E, Ichim M. Chemical composition and antioxidant activity of Thymus vulgaris L. volatile oil obtained by two different methods. ROM BIOTECH LETT, University of Bucharest. 2010; 15(4): 5436- 5443
4. Nanasombat S, Thonglong J, Jitlakha J. Formulation and characterization of novel functional beverages with antioxidant and anti-acetylcholinesterase activities. FFHD. 2015; 5(1): 1-16
5. Ocana A, Reglero G. Effects of Thyme extract oils (from Thymus vulgaris, Thymuszygis, and Thymus hyemalis) on cytokine production and gene expression of oxLDL-Stimulated THP-1Macrophages, J.Obes. 2012; 1-11, Doi:10.1155/2012/104706
6. Prasanth R, Ravi VK, Varsha PV, Satyam S. Review on Thymus vulgaris traditional uses and pharmacological properties. Med Aromat Plants. 2014; 3 (4):1-3.
7. Sharangi AB, Guha S. Wonders of leafy spices: Medicinal properties ensuring Human Health. Science International, 2003; 1(9):312-317.

Source of support: Nil, Conflict of interest: None Declared

[Copy right © 2020. This is an Open Access article distributed under the terms of the International Journal of Pharmaceutical Archive (IJPA), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.]