

Spectroscopic Analysis of Flavonoid Quercetin from Methanol Extracts of Emblica Officinalis Fruits

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ABSTRACT

A brown coloured flavonoid has been isolated from the column chromatography of methanol extracts of *Emblica officinalis* seeds. The structure of this brown powder was fully elucidated with the help of detailed spectroscopic study and chemical analysis like UV, IR, ¹H-NMR, ¹³C-NMR and MS. Quercetin is used for the treatment of allergies, cancer, cardiovascular diseases, inflammations and also act as antihistaminic compound. Quercetin, a flavonol, is one of the most potent antioxidants among polyphenols.

Keywords: *Emblica officinalis*, Methanol extracts, Brown powder, Flavonoid, Quercetin

I. INTRODUCTION

Emblica officinalis Gaertn. (*Phyllanthus emblica* Linn.), also known as amla, has been used in Ayurveda, the ancient Indian system of medicine. According to the main classic texts on Ayurveda, *Charak Samhita* and *Sushruta Samhita*, amla is regarded as the “best among rejuvenative herbs”, and the “best among the sour fruits” [1]. It belongs to family euphorbiaceae and is also known as Indian gooseberry [2]. The fruit also forms an important constituent of many Ayurvedic preparations such as *chyvanprash* and *triphala* and is regarded as “one of the best rejuvenating” herbs [3]. It is the richest source of antioxidants like vitamin C, emblicanin A and B, punigluconin, pedunculagin, catalase, glutathione peroxidase, tannin, trigalloyl, polyphenols, flavonoids, ellagic acid, phyllembic acid, gallic acid and tannic acids[4-6].

II. EXPERIMENTAL

2.1 General The melting point was determined on Lab fit melting point apparatus. A UV spectrum in ethanol was obtained on SHIMADZU UV-1800 UV spectrophotometer. An IR spectrum was recorded on SHIMADZU FTIR-8400S (Fourier Transforms infrared spectrophotometer). ¹H-NMR (400MHz) and ¹³C-NMR were recorded in MeOD on Bruker, Avance 400 MHz NMR spectrometer. Chemical shifts are given as δ with TMS as internal standard. A HR-mass spectrum was recorded on Agilent, 6540, Q-TOF (HR-MS) mass spectrometer.

2.2 Plant material Fruits of *Emblica officinalis* were purchased from a specific seed shop of Jammu’s district and classified systematically by Dr. Gurdev Singh of the botany department at Lovely Professional University.

2.3 Extraction and isolation The dried and crushed fruits (one kg) of *Emblica officinalis* were soaked in methanol for 120 hours. The crude extract of methanol was subjected to column chromatography and 5:3 Pet. ether: DCM fraction after keeping for around 2 months results into shiny, brown powder. The brown powder thus separated and recrystallised with ethanol thrice for the identification of secondary metabolite.

III. RESULTS AND DISCUSSION

The shiny brown powder obtained from methanol extracts of *Emblica officinalis* fruits was found to be a flavonoid on performing Shinoda test and Zinc Hydrochloride reduction test [7]. The flavonoid framework was also supported by UV and IR spectroscopy. The melting point was found out to be 315°C, which is very close to that of quercetin [8]. TLC of the powder which were recrystallised using ethanol showed Rf value equals to 0.99 that is similar to Rf value of quercetin observed in literature was 1.02 [9] thus powder obtained may be of quercetin.

3.1 The UV spectral Analysis UV Spectra peak observed at 368 nm is very close to 370 nm corresponds to flavonoid as shown in Fig. 1.

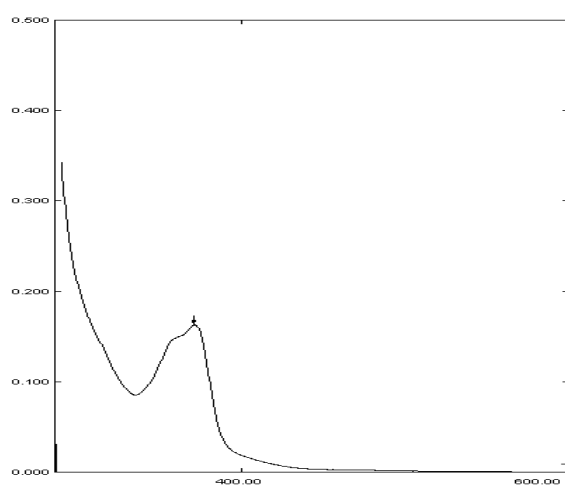


Fig. 1. UV spectrum of Brown powder from *Emblica officinalis* fruits

3.2 IR Spectral Analysis IR spectral peaks shows a band at 3564 cm^{-1} which is due to free O-H, at 3082 cm^{-1} showed the presence of intra molecular hydroxyl groups, 1618 cm^{-1} is due to carbonyl group (C=O), 1510, 1550, 1610 cm^{-1} showed the presence of C-C stretching of aromatic ring and 1111 cm^{-1} due to ether group as shown in Fig. 2.

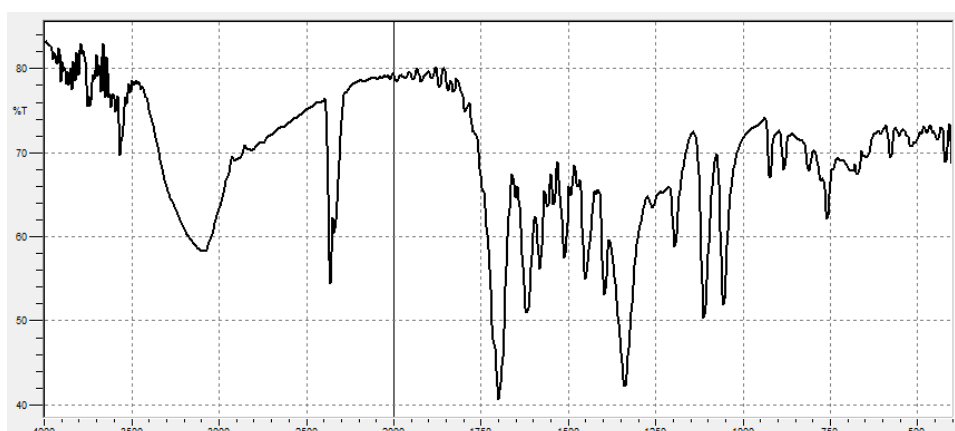


Fig 2. IR Spectra of brown powder

3.3 ¹H-NMR Spectral Analysis The ¹H-NMR (DMSO) shows values at δ 6.69(1H, d, J=1.7Hz), at δ 6.71(1H, d, J=1.7Hz) are due to meta-coupled protons of A-ring (H-6 and H-8) of a flavonoid nucleus. Signals at δ = 6.92 d, 7.4 and δ = 7.29 dd are assigned to H-5', H-2' and H-6' of the ring. Aromatic protons show various peaks in the region of 6 – 9.

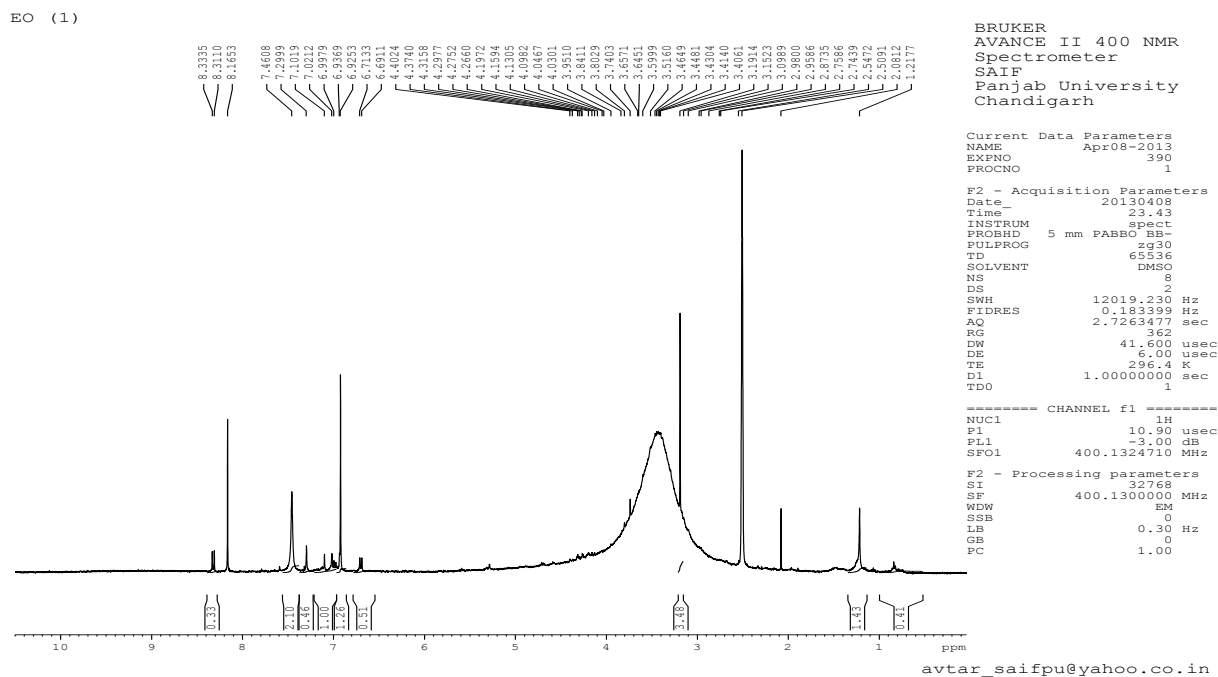


Fig. 3 ¹H-NMR Spectra

3.4 ¹³C-NMR Spectral Analysis is shown in Fig. 4.

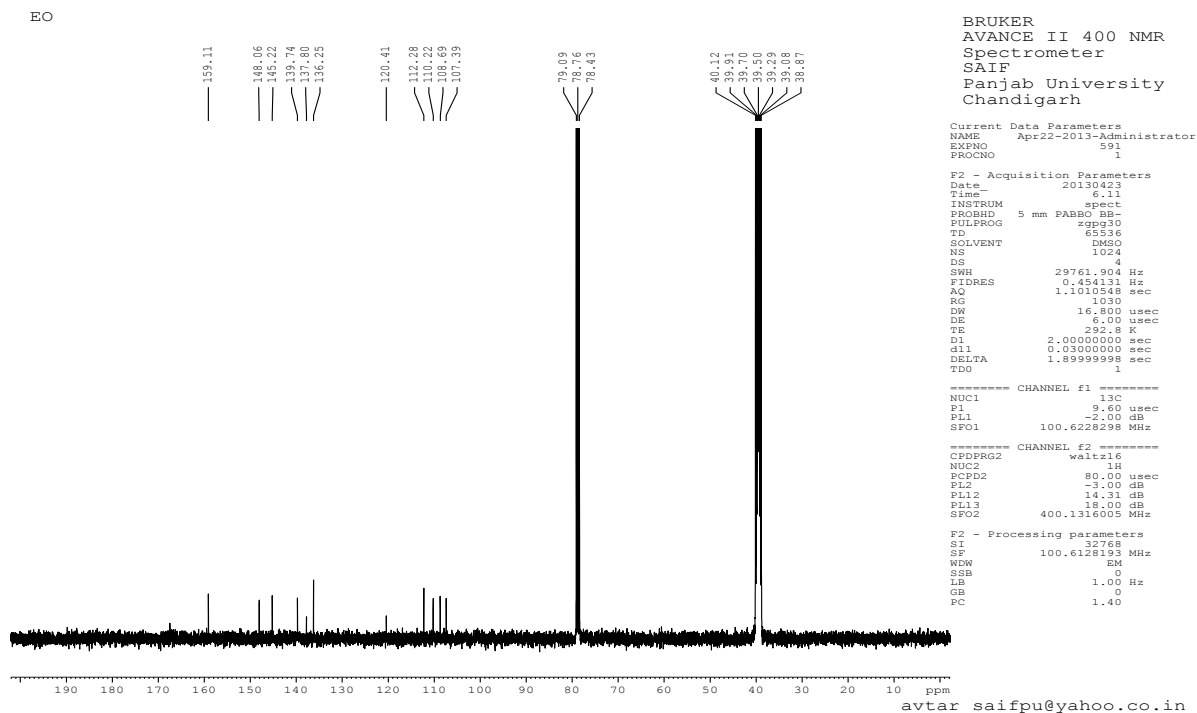


Fig. 4 ¹³C-NMR Spectrum

Justification of various peaks in ¹³C-NMR Spectrum

Position	¹³ C NMR
2	147.8
3	136.8
4	176.9
5	159.11
6	98.8
7	167.0
8	94.0
1'	120.41
2'	112.28
3'	145.22
4'	148.7
5'	116.2
6'	120.6

3.5 Mass Spectral Analysis: Mass Spectrum [10] of brown powder is shown below

Serial No.	Peaks	Justification of Peaks
1	301	[M-H] ⁻
2	302	[M+H] ⁺
3	303	[M+2H] ⁺
4	317	[M+OH+H] ⁺
5	300	[M-2H] ²⁻

IV. CONCLUSIONS

On the basis of above spectral analysis the brown powder may be classified as flavonol Quercetin as shown in Fig. 5.

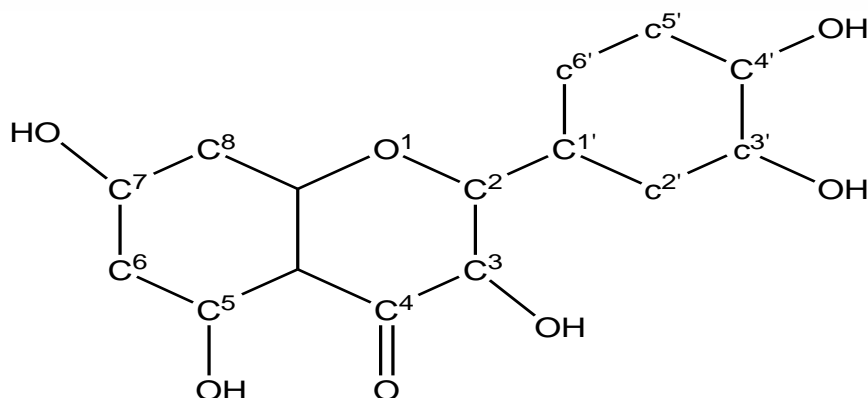


Fig. 5 Structure of Quercetin

V. ACKNOWLEDGEMENTS

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