

ANTIOXIDANT ACTIVITY, PRELIMINARY PHYTOCHEMICAL INVESTIGATION AND GC-MS STUDY OF BOUGAINVILLEA GLABRA CHOICY LEAVES

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ABSTRACT

The plant leaves contain a number of medicinally important compounds. The present study was carried out to identify the phytochemicals present in the Bougainvillea glabra leaves and evaluate antioxidant potential of the extract. Total phenol content was estimated by Folin Ciocalteu method and the phenolic content was 30.00mg/100 of gallic acid equivalent (GE). Antioxidant activity was evaluated by DPPH method and the leaves of Bougainvillea glabra showed 90.66mg/100 of Ascorbic acid Equivalent Antioxidant Capacity (AEAC). The GC-MS study also carried out and it showed the presence of phytochemicals like phytol,9,12,15-Octadecatrienoic acid (z,z,z) (RT:19.11), Squalene (RT:28.71) and Vitamins.

Keywords: Total phenols, Total flavonoids, Antioxidant activity, DPPH, GC-MS and Bougainvillea glabra .

INTRODUCTION

Bougainvillea glabra is a colourful flowers in front of houses and in office and on walls and fences. With origin from South America it is spread in the Caribbean and tropical regions^[1] and also grown in moderate climate regions ^[2, 3]. Bougainvillea glabra 'choicy' has been used in a variety of disorders including diarrhoea, acidity^[4]. Hence the present investigation was carried out to study the total flavonoids, phenolics, antioxidant activity of Bougainvillea glabra 'Choicy' leaves by GC-MS.

MATERIALS AND METHODS

Collection and processing of plant material

Leaves of the plant Bougainvillea glabra collected from Thanjavur District in the month of July, 2010 and authenticated by Dr. John Britto, Rapinet Herbarium, ST. Joseph's College, Tiruchirappalli.

The leaves were cleansed and shade dried for a week and grounded into uniform powder. 10g of plant material was added to 200 ml of aqueous ethanol (20%,v/v) for 18 hours at room temperature. Filtered extracts were used for the estimation of preliminary phytochemical studies and antioxidant activity.

Phytochemical analysis

Phytochemical analysis involves the qualitative and quantitative analysis of herbal plants. The preliminary qualitative and quantitative tests have been attempted in Bougainvillea glabra leaves to find out the presence or absence of certain bioactive compounds.

Phytochemical Screening

The preliminary screening was carried out by using standard procedures described by Sofowara ^[9], Treese and Evans ^[10] and Harborne ^[11].

Quantitative determination of the chemical constituents

Determination of total phenols

Total phenol content was determined using standard procedure^[12].

Determination of total flavonoids

Quantification of total flavonoids was done using published protocols ^[13].

Antioxidant activity

Antioxidant capacity was determined using known methods^[14].

GC-MS analysis

Preparation of extract

Leaves of Bougainvillea glabra were shade dried. 20g of the powdered leaves were soaked in 95% ethanol for 12 hrs. Sediments were removed and concentrated by bubbling nitrogen gas into the solution. The extract contained both polar and non-polar phytochemicals of the plant material used. 2µl of these solutions was employed for GC-MS analysis^[15].

GC analysis

GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument using reported conditions.

Identification of components

Interpretation on mass spectrum GC-MS was conducted using the database (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum.

Table 1: Qualitative analysis of the phytochemicals of Bougainvillea glabra

S. No	Phytochemical	Result
1	Tannins	Present
2	Phlobatannins	Present
3	Saponin	Present
4	Flavonoids	Present
5	Steroids	Present
6	Terpenoids	Present
7	Cardiac glycosides	Present

Table 2: Total phenolics, flavonoids and antioxidant activity in the leaves of Bougainvillea glabra

S. No	Parameter analysed	Values obtained
1	Total phenols (mg/100g) GE*	30.00 ± 0.8
2	Total flavonoids (mg/100g) GE*	0.40 ± 0.3
3	Antioxidant activity (mg/100g) AEAC**	90.66 ± 0.94

The values are mean value of three replicates.* Gallic acid equivalent, **Ascorbic acid equivalent antioxidant capacity

Table 3: Phytochemicals identified in the ethanolic extract of the leaves of Bougainvillea glabra by GC-MS

	Name of the compound	Peak area(%)
12.95	3-O-Methyl-d-glucose	77.04
16.48	Tetradecanoic acid, ethyl ester	1.38
18.36	Phytol	6.58
19.11	9,12,15-Octadecatrienoic acid, (Z,Z,Z)	1.23
22.44	Hexanedioic acid. bis (2-ethylhexyl) ester	0.94
24.59	1,2-Benzenedicarboxylic acid, diisooctyl ester	9.91
28.71	Squalene	1.94
35.71	Vitamin E	0.98

Table 4: Activity of phytochemicals identified in Bougainvillea glabra extract by GC-MS

No	RT	Name of the compound	Molecular Formula	MW	Peak Area%	Compound Nature	**Activity
1	12.95	3-O-Methyl-d-glucose	C ₇ H ₁₄ O ₆	194	77.04	Sugar moiety	Preservative
2	16.48	Tetradecanoic acid, ethyl ester	C ₁₆ H ₃₂ O ₂	256	1.38	Fatty acid ester	Antioxidant, Cancer preventive, Nematicide, Lubricant
3	18.36	Phytol	C ₂₀ H ₄₀ O	296	6.58	Diterpene	Hypocholesterolemic
4	19.11	9,12,15-Octadecatrienoic acid, (Z,Z,Z)	C ₁₈ H ₃₀ O ₂	278	1.23	Linolenic acid ester	Anticancer, Antiinflammatory, Antioxidant, Diuretic
5	22.44	Hexanedioic acid. bis (2-ethylhexyl) ester	C ₂₂ H ₄₂ O ₄	370	0.94	Ester compound	Hypocholesterolemic, Nematicide, Antiarthritic, Hepatoprotective, Anti androgenic
6	24.59	1,2-Benzenedicarboxylic acid, diisooctyl ester	C ₂₄ H ₃₈ O ₄	390	9.91	Plasticizer compound	Hypocholesterolemic, 5-Alpha reductase inhibitor, Antihistaminic, Anticoronary, Insectifuge, Antieczemic, Antiacne
7	28.71	Squalene	C ₃₀ H ₅₀	410	1.94	Triterpene	No activity reported
8	35.71	Vitamin E	C ₂₉ H ₅₀ O ₂	430	0.98	Vitamin compound	Antimicrobial, Antifouling, Antibacterial, Antioxidant, Antitumor, Cancer preventive, Immunostimulant, Chemo preventive, Lipoxygenase-inhibitor, Pesticide, Antiageing, Analgesic, Antidiabetic, Antiinflammatory, Antioxidant, Antidermatitic, Antileukemic, Antitumor, Anticancer, Hepatoprotective, Hypocholesterolemic, Antiulcerogenic, Vasodilator, Antispasmodic, .Antibronchitic, Anticoronary

**Source: - Dr. Duke's Phytochemical and Ethnobotanical Databases

Bougainvillea glabra leaf 152

GC-MS Analysis 106

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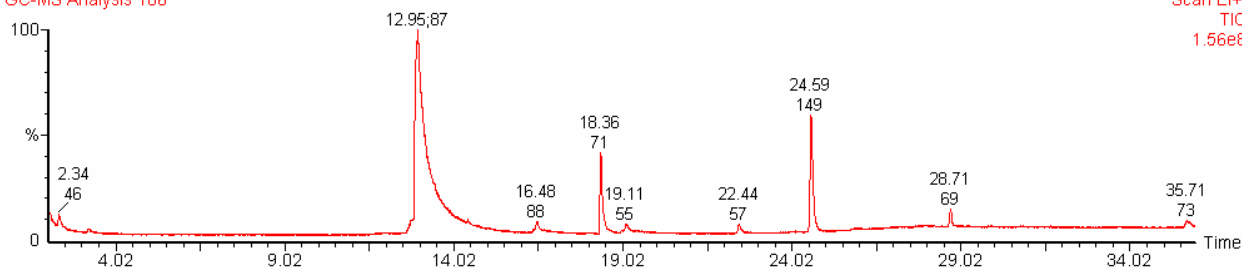


Fig. 1: GC MS Chromatogram of Bougainvillea glabra 'Choicy'

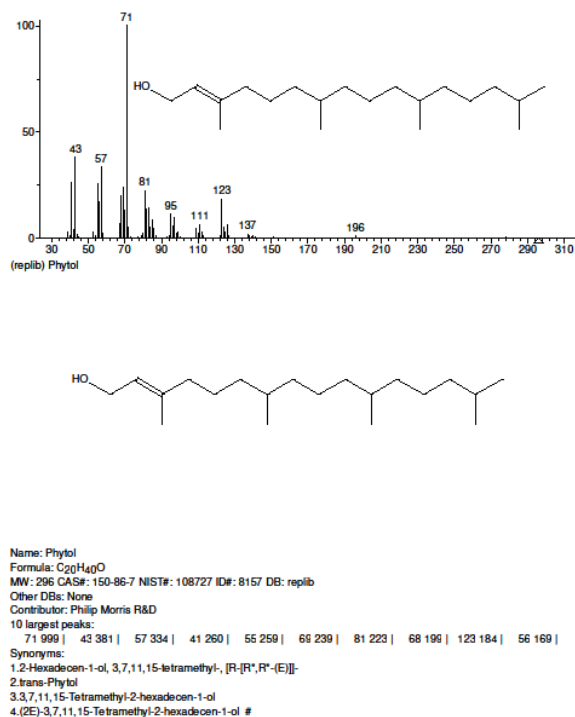


Fig. 2: mass spectrum and structure of phytol

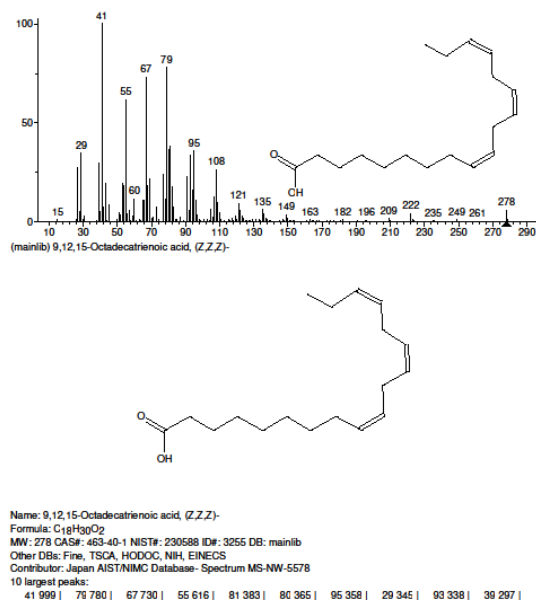


Fig. 3: mass spectrum and structure of 9, 12, 15-Octadecatrienoic acid (z,z,z).

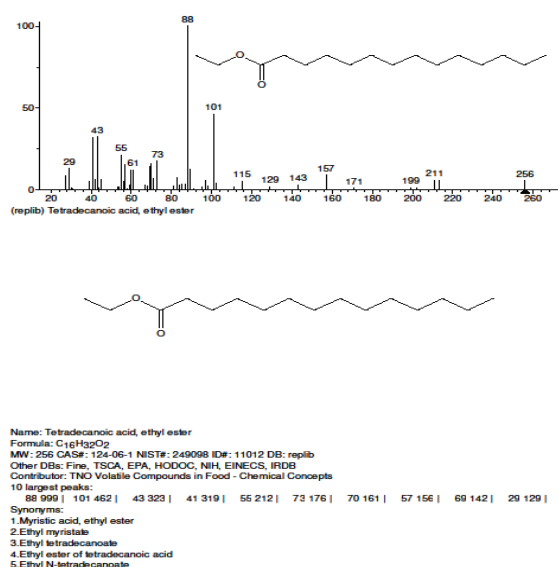


Fig. 4: Mass spectrum and structure of Tetradecanoic acid, ethyl ester.

RESULTS AND DISCUSSION

Qualitative and Quantitative analysis of Bougainvillea glabra leaves

The phytochemical characters of the Bougainvillea glabra leaves investigated are summarized in (Table 1). Tannins, flavonoids, saponin, steroid, phlobatanin, terpenoid and cardiac glycosides were present in Bougainvillea leaves.

Flavonoids are known to elicit antioxidant activity and find use in human nutrition and health [16]. Total phenolic content of the ethanolic extract of Bougainvillea leaves (Table 2) is 30.00 mg/100 g of GE. The value of phenolic content indicates that the plant has high antioxidant activity [17].

GC-MS Study

The GC-MS study of Bougainvillea leaves has shown many phytochemicals which contribute to the medicinal activity of the plant (Tables 3 and 4). The major components which present 3-O-Methyl-d-glucose, Tetradecanoic acid, ethyl ester, Phytol, 9,12,15-Octadecatrienoic acid, 1,2-Benzenedicarboxylic acid, diisooctyl ester, Squalene. The other compounds like Hexanedioic acid, Vitamin E present in the leaves of Bougainvillea (figure 1). Figure 2,3, and 4 shows mass spectrum and structure of phytol, 9,12,15-Octadecatrienoic acid and coumarin, Tetradecanoic acid, ethyl ester compound which is suggested to be a diterpenoid, Linolenic acid ester and Fatty acid ester compound and is used as an anticancer, anti-inflammatory, antioxidant, antimicrobial, diuretic, Nematicide, Anticoronary, Insectifuge and Antieczemic.

CONCLUSION

The study clearly indicates that the leaf extract was high in antioxidants, phenolics and normal in flavonoids. The GC-MS study also showed many Phytochemicals 3-O-Methyl-d-glucose, Tetradecanoic acid, ethyl ester, Phytol, 9,12,15-Octadecatrienoic acid, Hexanedioic acid, 1,2-Benzenedicarboxylic acid, diisooctyl ester, Squalene, Vitamin E, phytol, Squalene which contribute to the activities like antimicrobial, antioxidant, anticancer, hypercholesterolemic, antiulcerogenic and other activities. This

investigation has identified the compounds present in the leaves of Bougainvillea, and evaluation of pharmacological activity in the ethanol extract is in progress.

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Abbreviations: GE, Gallic acid equivalent; DPPH, 2,2-diphenyl-1-picrylhydrazyl; AEAC, ascorbic acid equivalent antioxidant capacity; GC-MS, gas chromatograph and mass spectrometer, UV, ultraviolet; NIST, National Institute Standard and Technology; O.D, optical density.

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