

GC-MS DETERMINATION OF BIOACTIVE COMPONENTS OF *POLYGALA JAVANA* DCM. ALAGAMMAL¹, P.S. TRESINA² AND V.R. MOHAN^{2*}¹Government Siddha Medical College, Palayamkottai, Tamil Nadu, ²Ethnopharmacology Unit, Research Department of Botany, V.O.Chidambaram College, Tuticorin-628008, Tamil Nadu, India. Email: vrmohan_2005@yahoo.com

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ABSTRACT

In this study, the bioactive components of *Polygala javana* whole plant have been evaluated using GC-MS. The chemical compositions of the ethanol extract of *Polygala javana* were investigated using Perkin-Elmer Gas Chromatography-Mass Spectrometry, while the mass spectra of the compounds found in the extract was matched with the National Institute of Standards and Technology (NIST) library. GC-MS analysis of ethanol extract of *Polygala javana* revealed the existence of Polygalitol (84.79%), 1H-Perimidine, 2,3-dihydro-2-(2,4,5-trimethoxyphenyl) (6.33%), 4H-1-Benzopyran-4-one, 5-hydroxy-2-(4-hydroxyphenyl)-3,7-dimethoxy (1.53%) and Ledene oxide-(1) (1.43%). This is the first report of identification of active constituents from whole plant of *Polygala javana* by GC-MS.

Keywords: *Polygala javana*, Ledene oxide (1), Squalene, Anti-cancer

INTRODUCTION

Genus *Polygala* is an annual, diffuse herb, 10-25cm tall. Flowers are papilionaceous, primary root orange, stems woody at base, branches terate, crisped pubescent. Leaf blade green, obovate, elliptic or lanceolate, 2.6-10x1-1.5cm, papery, pubescent, inflorescence raceme, super-axillary, rarely axillary, shorter than leaves, densely few flowered. Flowers 4-5mm long, sepals 5, persistent, green, ciliate, outer sepals 3, ovate-lanceolate, apex acuminate; inner sepals 2, petaloid, falcate, petals 3, connate at base, yellowish or white with pink, lateral petals shorter than keel, inside with fasciated white hairs at base. Stamens 8, ovary compressed- orbicular. *Polygala* was traditionally used by Americans to treat snake bites¹ and as an expectorant to treat cough and bronchitis. *Polygala* is considered as a powerful tonic herb² that can help to develop the mind and aid in creative thinking.

Polygala javana DC belongs to Polygalaceae family. It is commonly known as "Palpiranthai". Paste prepared from fresh leaves is applied by Kanikkar tribal women on the breast twice a day for 2-3 days to check lactation and to get relief from the pain developed while stopping mother feeding³. Taking into consideration of the medicinal importance of *Polygala javana*, the ethanol extract of whole plant of *Polygala javana* were analyzed for the first time using GC-MS. This work will help to identify the compounds of therapeutic value.

MATERIALS AND METHODS

Materials and Methods

Collection of plant sample

Whole plant of *Polygala javana* was collected from Courtallam, Tirunelveli District, Tamilnadu. With the help of local flora, voucher specimens were identified and presented in the Ethnopharmacology Unit, Research Department of Botany, V.O.Chidambaram College, Tuticorin, Tamil Nadu for further references.

Plant sample extraction

Whole plants were cleaned, shade dried and pulverized to powder in a mechanical grinder. Required quantity of powder was weighed and transferred to Stoppard flask, and treated with ethanol until the powder is fully immersed. The flask was shaken every hour for the first 6 hrs and then it was kept aside and again shaken after 24 hrs. This process was repeated for 3 days and then the extract was filtered. The extract was collected and evaporated to dryness by using vacuum distillation unit. The final residue thus obtained was then subjected to GC-MS analysis.

GC-MS analysis

GC-MS analysis of these extracts was carried out by following the method of Hema *et al*⁴. GC-MS analysis were performed using a

Perkin-Elmer GC clauses 500 system and Gas chromatograph interfaced to a Mass spectrometer (GC-MS) equipped with a Elite-I, fused silica capillary column(30m x 0.25 mm ID x 1 μ df), composed of 100% Dimethyl poly siloxane). For GC/MS detection, an electron ionization system with ionizing energy of 70 eV was used. Helium gas (99.999%) was used as the carrier gas at constant flow rate 1ml/min and an injection volume of 2 μ l was employed split ratio of 10:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min) with an increase of 10°C / min to 200°C, then 5°C/min to 280°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5 seconds and fragments from 45 to 450 Da. Total GC running time was 36 minutes. The relative % amount of each component was calculated by comparing its average peak area to the total areas, software adopted to handle mass spectra and chromatograms was a Turbomass.

Identification of components

Interpretation on mass spectrum GC-MS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The Name, Molecular weight and Structure of the components of the test materials were ascertained.

RESULTS AND DISCUSSION

The compounds present in the ethanol extract of whole plant of *Polygala javana* were identified by GC-MS analysis (Fig. 1). The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration % in the ethanol extract of whole plant of *Polygala javana* are presented in Table 1. The prevailing compounds in ethanol extract of whole plant were Polygalitol (84.79%), 1H, Perimidine, 2,3-dihydro-2-(2,4,5-trimethoxyphenyl) (6.33%), 4H-1-Benzopyran-4-one, 5-hydroxy-2-(4-hydroxyphenyl)-3,7-dimethoxy- (1.53%), Ledene oxide- (1) and phytol (1.28%). Figures 2, 3 and 4 shows mass spectrum and structures of squalene and phytol. Table 2 listed the major phytocomponents and its biological activities obtained through GC-MS study of *Polygala javana*. Among the identified phytochemicals (Table 1) squalene have the property of antioxidant⁵. Recently squalene possesses chemopreventive activity against colon carcinogenesis⁶. Phytol is detected in *Polygala javana* whole plant which was also found to be effective at different stages of the arthritis. It was found to give food as well as preventive and therapeutic results against arthritis. The results show that, reactive oxygen species-promoting substances such as phytol constitute a promising novel class of pharmaceuticals for the treatment of rheumatic arthritis and possibly other chronic inflammatory diseases⁷.

In the present study, sixteen chemical constituents have been identified from ethanol extract of the whole plant of *Polygala javana* by GC-MS analysis. The presence of various bioactive compounds justifies the use of the whole plant for various

ailments by traditional practitioners. So it is recommended as a plant of phytopharmaceutical importance. However, further studies will need to be undertaken to ascertain fully its bioactivity.

Table 1: Components detected in the whole plant ethanol extract of *Polygala javana*.

Sr. No.	RT	Name of the compound	Molecular Formula	MW	Peak Area %
1.	2.07	Butane, 1,1-diethoxy-2-methyl-	C ₉ H ₂₀ O ₂	160	0.51
2.	2.29	Glycerin	C ₃ H ₈ O ₃	92	0.58
3.	2.88	Propane, 1,1,3-triethoxy-	C ₉ H ₂₀ O ₃	176	0.24
4.	11.27	Polygalitol	C ₆ H ₁₂ O ₅	164	84.79
5.	11.66	1,14-Tetradecanediol	C ₁₄ H ₃₀ O ₂	230	0.42
6.	13.10	Dibutyl phthalate	C ₁₆ H ₂₂ O ₄	278	0.11
7.	13.63	2-Pentanone, 1-(2,4,6-trihydroxyphenyl)	C ₁₁ H ₁₄ O ₄	210	0.17
8.	15.00	Phytol	C ₂₀ H ₄₀ O	296	1.28
9.	16.14	Ledene oxide-(I)	C ₁₅ H ₂₄ O	220	1.43
10.	18.88	Cedran-diol, 8S,14-	C ₁₅ H ₂₆ O ₂	238	0.35
11.	20.87	Didodecyl phthalate	C ₃₂ H ₅₄ O ₄	502	0.10
12.	21.88	4,8,12,16-Octadecatetraen-1-ol, 4,9,13,17-tetramethyl-	C ₂₂ H ₃₈ O	318	0.78
13.	24.71	Squalene	C ₃₀ H ₅₀	410	0.87
14.	28.00	4H-1-Benzopyran-4-one, 5-hydroxy-2-(4-hydroxyphenyl)-3,7-dimethoxy-	C ₁₇ H ₁₄ O ₆	314	1.53
15.	32.33	Spiro[androst-5-ene-17,1'-cyclobutan]-2'-one, 3-hydroxy-, (3 α ,17 α)-	C ₂₂ H ₃₂ O ₂	328	0.49
16.	34.95	1H-Perimidine, 2,3-dihydro-2-(2,4,5-trimethoxyphenyl)-	C ₂₀ H ₂₀ N ₂ O ₃	336	6.33

Table 2: Activity of phyto-components identified in the ethanolic extracts of the whole plant of *Polygala javana* by GC-MS

Sr. No.	RT	Name of the compound	Molecular Formula	Nature of compound	**Activity
1	2.29	Glycerin	C ₃ H ₈ O ₃	Alcoholic compound	Antimicrobial Preservative
2	11.66	1,14-Tetradecanediol	C ₁₄ H ₃₀ O ₂	Alcoholic compound	Antimicrobial
3	13.10	Dibutyl phthalate	C ₁₆ H ₂₂ O ₄	Plasticizer compound	Antimicrobial Antifouling
4	15.00	Phytol	C ₂₀ H ₄₀ O	Diterpene	Antimicrobial Antiinflammatory Anticancer Diuretic
5	16.14	Ledene oxide-(I)	C ₁₅ H ₂₄ O	Sesquiterpene oxide	Anti-tumor, Analgesic Antibacterial, Antiinflammatory Sedative, Fungicide
6	18.88	Cedran-diol, 8S,14-	C ₁₅ H ₂₆ O ₂	Sesquiterpene alcohol	Antimicrobial Antiinflammatory
7	20.87	Didodecyl phthalate	C ₃₂ H ₅₄ O ₄	Plasticizer compound	Antimicrobial Antifouling
8	21.88	4,8,12,16-Octadecatetraen-1-ol, 4,9,13,17-tetramethyl-	C ₂₂ H ₃₈ O	Unsaturated alcohol compound	Antimicrobial
9	24.71	Squalene	C ₃₀ H ₅₀	Triterpene	Antibacterial, Antioxidant, Antitumor, Cancer preventive, Immunostimulant, Chemo preventive, Lipoygenase-inhibitor, Pesticide Diuretic
10	34.95	1H-Perimidine, 2,3-dihydro-2-(2,4,5-trimethoxyphenyl)-	C ₂₀ H ₂₀ N ₂ O ₃	Alkaloid compound	Antimicrobial Antioxidant Antiinflammatory

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

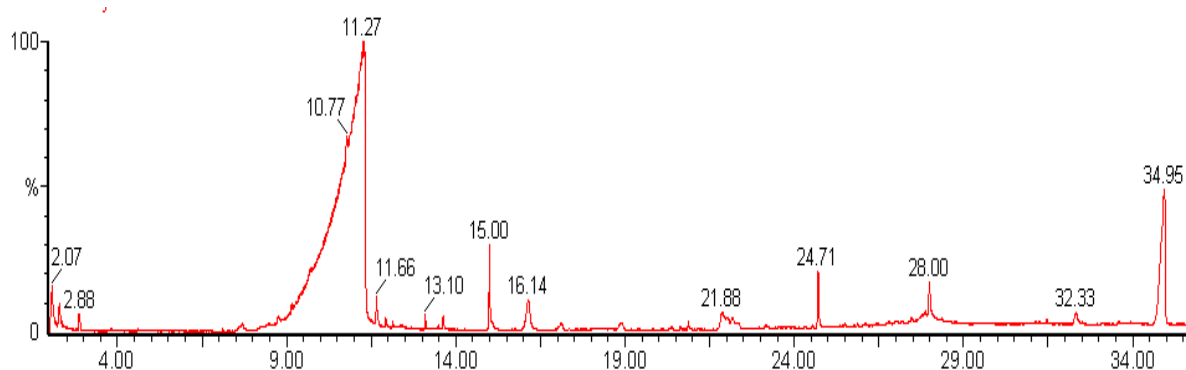


Fig. 1: GC-MS chromatogram of the ethanolic extract of the whole plant of *Polygala javana*

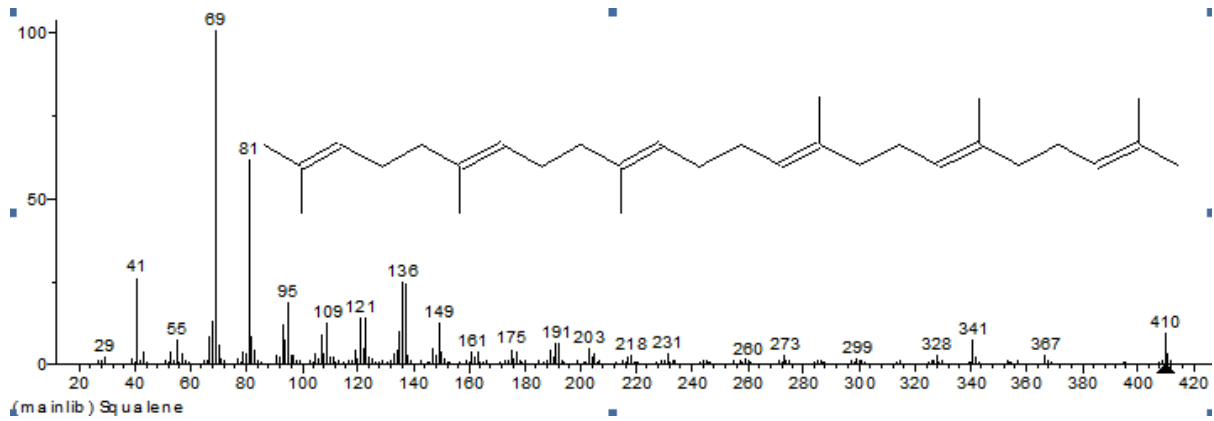


Fig. 2: Mass spectrum of Squalene

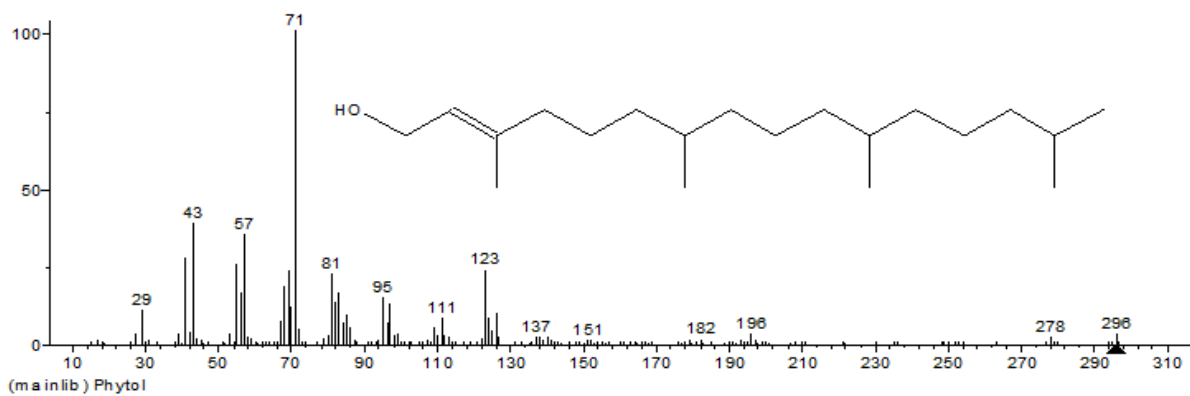


Fig. 3: Mass spectrum of phytol

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