

GC-MS analysis of *Gymnema sylvestre* leaves methanolic extract for antidiabetic and anticancer drug identification

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ABSTRACT

Methanol extraction has applied to *Gymnema sylvestre* leaves using soxhlet device for the determination of bioactive compounds was our primary aim. Extraction from the process was analysed by GC-MS. Using standard reference from NIST, finding the existence of 7 phyto chemical compounds with anti- diabetic, cancer activities. The major phyto constituents were 9 octo decanoic acid (Z), methyl ester (12.93), n hexa decanoic acid [12.08] [E], 9octo decanoic acid ethyl ester (13.66), 9, 12 octo decanoic acid (Z, Z) (14.35), 1,3-propanediol, 2-ethyl-2 (hydroxymethyl) -(Ethriol) (11.56) [1,1'-Bicyclopropyl]-2-octanoic acid, 2'-hexyl-,methyl ester (12.88), Squalene (21.85). Phyto chemical analysis were made for the extracts to confirm the phyto chemicals existing in the leaves. The biological activities are based on pass online Databases. Production and identification of anticancer and anti-diabetic compound helps to approach herbal as an alternate remedy.

KEY WORDS: *Gymnema Sylvestre*, Soxhlet extraction, GC-MS, Anti-diabetic and Anticancer activity.

1. INTRODUCTION

Gymnema Sylvestre belonging to the family Asclepiadaceae grows in open forest of one division in our country and Asia (Wu, 2012). It has a fame in folk medicine as a stomachic, diuretic and it cure to control diabetes mellitus. Its application in the area of asthma, snakebites, eye ailments, chronic cough, breathing troubles and also to assist in family planning (Patel, 2012). Basic metabolic tests of leaves shows, they are 2.5 - 6 cm long and usually elliptical. The flowers are small, yellow, and it umbellate cymes, and the cavities are terete, lanceolate and upto 7.5 cm in length (Kanetkar, 2007). The presence of flavinoids, quinines, phenolic acids, saponins has been recorded in the leaves of *Gymnema Sylvestre* (Thakur, 2012). The leaves of *Gymnema Sylvestre* lower blood sugar, stimulate the heart, uterus, cardiovascular, and exhibit anti-sweet and hepato protectice activities (Kang, 2012; Baskaran, 1990; Ahmed, 2010)

2. MATERIAL AND METHODS

Collection and processing of plant material: The leaves of *Gymnema sylvestre* were collected from the natural livings of Kanchipuram district, Tamil Nadu, India. Inorder to clean the gems, after cleaning. The Leaves were cut, shade dried, ground into fine powder using mechanical grinder and it could be kept in a permissible safety material. **Plant sample extraction:** 250grams of powdered material was located inside a thimble made from thick filter paper, loaded into the main chamber of the soxhlet extractor. 2500ml of Methanol is also loaded into a chamber and flow in a condenser. Methanol was heated to reflux. Methanol flows into part of an instrument like conical arm allows a system using a solid. The device allowed to methanol vapour cools and drips back down. When the devices is completed level of methanol, in eight hours. Meanwhile methanol completed in full swing, solved and easily vapourized. After extraction the solvent is removed by rotary evaporator, and extracts collected for analyzing GC-MS.

GC-MS analysis: Extracts were performed in IICPT, Tanjore, Tamilnadu. 2 μ l of the solvent extract of *Gymnema sylvestre* was employed. The device performed in the analysis as a fused capillary column packed with Elite-5MS [5%diphenyl, 95% dimethyl polysiloxane] 30 mm \times 0.25 mm ID \times 0.25 μ m df]; and the device were divided by Helium at a steady flow of 1 ml/min. Specimen were allowed to the device was found with 5.2 software. During the 36thminute this reaction was kept at 280 $^{\circ}$ C by keeping 720sec. Initially we set at 250 $^{\circ}$ C. Various criteria as included at 500 μ s. Mass spectra were taken at 70 eV in between 0-2 minutes and components ranges from 45 to 450 Da. The process was completed in 36minutes.

Preliminary phyto chemical screening: Freshly prepared extracts were allowed to a standard phyto chemical analysis to ensure major chemical components by standard descriptions.

PASS computer program: Prediction of solvent extract of *Gymnema sylvestre* leaves for anti-diabetic and anti-cancer activity was done with the help of computer program.

PASS is a computer based program for different substances including phyto constituents. Prediction of this spectrum by PASS is based on structural activity relationship (SAR). In this area 2,05,000 compounds exhibiting greater number of research activities. Higher assumption of a compound is estimate as probable activity (Pi). The compounds showing more Pa value than Pi are the only way for the bio activity.

Identification of compounds: Compounds were identified in area by bit by bit through NIST ver 2.0 year 2005 library.

3. RESULTS

Phytochemical prospective: Solvent extract of *Gymnema sylvestre* leaves showed presence of flavinoids, quinines, phenolic acids, and saponins.

PASS predictions for Antidiabetic and Anticancer: Our current research work predictions were identified and applied. PASS predicted probable activity (Pa) of Sugar-phosphatase, Sphinganine kinase for Antidiabetic and Anticancer activity was 0.904 & 0.04 and 0.924 & 0.004 respectively which is greater than 0.7. This type of probable activity spectra was not shown by other phytochemicals from *Gymnema sylvestre* leave extract.

Discussion: Preliminary phytochemical investigation confirmed presence of Alkaloid, saponins, phenolic compounds and tannins which establishes richness of plant in secondary metabolites.

The results find outs, what are all the compounds available from our work patterns. It will found out through chromatogram. Recorded values from the methanol extraction proves for the responsible for antibacterial activity.

GC-MS analysis was done using the organic solvent methanol and it showed the presence of 7 different chemical compounds present in the plant sample vide Table1, Fig.1. The sample was extracted with methanol because of the effect of antidiabetic and anticancer activity in this solvent. GC-MS analysis also provides the spectrum for the methanolic extract. From the chromatogram shows the presence of seven components with the retention time 11.56 & 21.85 respectively.

The biological activities based on Pass online databases were tabulated in Table 2. The phyto chemical screening chemical constituents of the plants studied showed that the leaves were amino acids and secondary metabolites such as alkaloids, flavonoids. They have both therapeutic and physiological value

GC-MS exhibit *Gymnema sylvestre* leaves of relative concentrations of various compounds with a function of retention time were illustrated in the graph.

Table.1. Identification of compounds from GC-MS

RT	Name of the compound	Molecular Formulae	MW	Peak Area %	Structure
11.56	1,3-Propanediol, 2-ethyl-2-(hydroxymethyl)- (Ethriol)	C ₆ H ₁₄ O ₃	134	8.87	
12.08	n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256	15.56	
12.88	[1,1'-Bicyclopropyl]-2-octanoic acid, 2'- hexyl-, methyl ester	C ₂₁ H ₃₈ O ₂	322	1.53	
12.93	9-Octadecenoic acid (Z)-, methyl ester	C ₁₉ H ₃₆ O ₂	296	1.50	
13.66	(E)-9-Octadecenoic acid ethyl ester	C ₂₀ H ₃₈ O ₂	310	0.25	
14.35	9,12-Octadecadienoic acid (Z,Z)-	C ₁₈ H ₃₂ O ₂	280	72.25	
21.85	Squalene	C ₃₀ H ₅₀	410	0.05	

Table.2. Biological activity of 1,3-Propanediol, 2-ethyl-2-(hydroxymethyl)- (Ethriol) [C₆H₁₄O₃]

Pa	Pi	Compound predicted
0,932	0,002	Alkyl acetyl glycerol phosphatase inhibitor
0,924	0,004	Sphinganine kinase inhibitor
0,913	0,004	Ubiquinol-cytochrome-c reductase inhibitor
0,911	0,003	Fucosterol-epoxide lyase inhibitor
0,910	0,004	Alkenyl glycerol phosphocholine hydrolase inhibitor
0,904	0,004	Sugar-phosphatase inhibitor
0,902	0,005	Acrocylindro pepsin inhibitor
0,902	0,005	Chymosin inhibitor
0,902	0,005	Saccharo pepsin inhibitor
0,894	0,013	CYP2C12 substrate

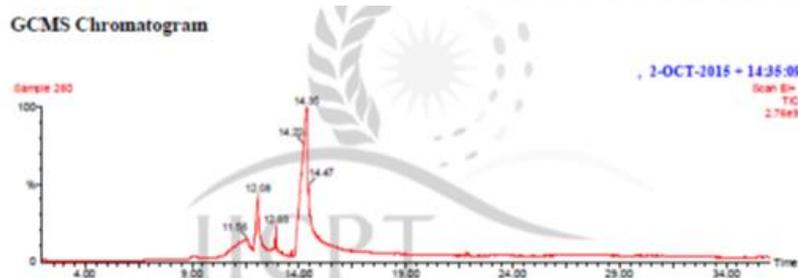


Figure.1. GC-MS Chromatogram

4. CONCLUSION

Thus the findings of present investigation support the traditional ethno medicinal claims of *Gymnema sylvestre* for the treatment of diverse infections. Identified phyto components using GC-MS can be used as a commercial device for the identification of PASS online biological activity predicts. It opens the way to evolve different treatment regimens used this extract. Further studies are required to need to treat this plant material is an effective anti-diabetic and anticancer agent.

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