



## GC-MS ANALYSIS OF ESTER AND ETHANOL EXTRACTS OF *Stevia Rebaudiana* (LEAVES)

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### ABSTRACT

Abstract: Natural products have played a very important role in health care and prevention of diseases. The investigation was carried out to determine the chemical constituents of the ester and extract of *Stevia rebaudiana* using GC-MS. GC-MS analysis of the ester and ethanol extract led to identification of 13 and 10 compounds. The compounds found in ester extract are Acetophenone, 4-hydroxy-3, 5-dimethoxy, oxime, bis-TMS, 7-Octadecyne, Butyl tridecyl ether, Succinic acid, cyclohexylmethyl 2-pentyl ester, Cyclobutanecarboxamide, N-decyl-N-methyl, 1,2-Benzene dicarboxylic acid, dibutyl ester, Decane, Undecane, 1-Hexanamine, N-isopropylidene, 9-Octadecenamide. The compounds were identified by comparing their Retention time and covate indexes with that of literature and by interpretation of mass spectra. The compounds found in ethanol extract are Benzene, 1,3-dimethyl, 1-Hexene, 2,4,6-tris-trimethylsilyl, Methyl farnesoate, Hexadecanoic acid methyl ester, Pentadecanoic acid methyl ester, Nonyl pentyl ether, 1,2-Benzene dicarboxylic acid, dibutyl ester, Tricosanol, Heptacosane, Nonacosane and Octacosane.

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### INTRODUCTION

Natural products research continues to explore a variety of lead structures, which may be used as templates for the development of new drugs by the pharmaceutical industry. The most common high intensity sweeteners are the synthetic compounds such as aspartame, neotame and saccharin. Saccharin is associated with the potential risk of cancer of bladder when used in excess. Consumption of aspartame leads to metabolic disease phenyl-ketouria. Therefore, low calorie sweeteners have been investigated to substitute sugar. *Stevia* leaf is used for sweetening food and contains several sweet glycosides [1] [2] [3]. Most research has focused on the economically important species *stevia*. *Stevia rebaudiana* leaves used as a sweetener, antiseptic, healing tonic, stomachic, anthelmintic. *Stevia rebaudiana* leaves contain lactones, terpenoids, steroids, flavanoids, protein, carbohydrate, minerals, tannins [4] [5]. In the present paper, we report the phytochemicals isolated from ester and ethanol extract by gas chromatography mass spectrometry (GC-MS).

### MATERIALS AND METHODS

#### Plant material and Extract preparation

The dried leaves of *S. rebaudiana* were purchased from Sun fruit Pvt. Ltd Pune.

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The leaves of *S. rebaudiana* were shade dried, powdered and extracted in soxhlet extractor serially with n-hexane, dichloro methane, ethyl acetate, acetone, methanol, ethanol and water. Removal of solvent under reduced pressure afforded solid extracts. The removal of solvent under reduced pressure by rotary film evaporator yielded 6gm of ester and 4 gm of ethanol extract. As the yield of ester and ethanol extract is not good so it is not separated by column chromatography and analysed by GC-MS.

#### GC-MS analysis

Instrument and chromatographic conditions: Nowadays GC-Mass spectrometers are widely used to identify unknown compounds by way of determining their molecular mass and molecular formula at the expense of negligible amount of sample. Mass spectrometry analysis was performed on Shimadzu GCMS-QP-2010 SE model using Direct Injection Probe technique.

#### Identification of compounds

Compounds were identified by comparing mass spectra data of samples with those of the NIST (National Institute of Standards and Technology, USA) standard reference database. The quantitative estimation of each peak obtained in GC was made by computer, attached with GC-MS instrument. Literature reports already published, also helped in understanding the structure, as well as by comparison of the fragmentation patterns of the compounds present in *Stevia rebaudiana* [6] [7] [8] [9].

Phytochemicals identified in ester extracts of *S. rebaudiana* leaves (RS-2)

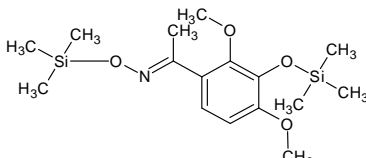
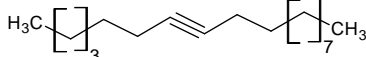
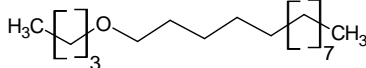
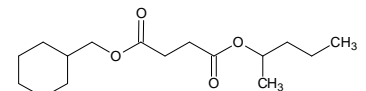
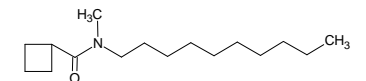
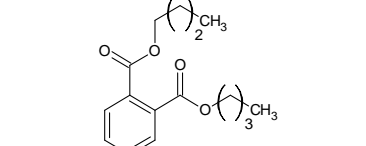
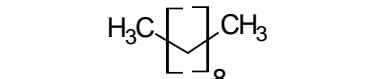
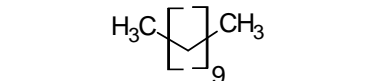
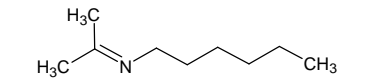
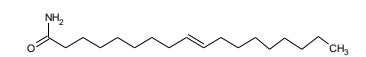
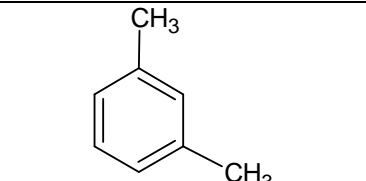
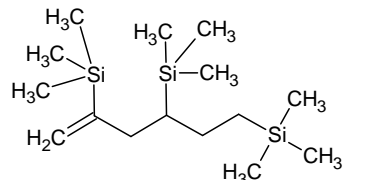
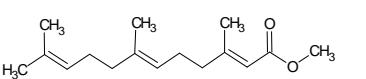
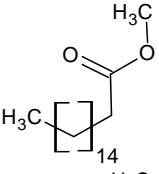
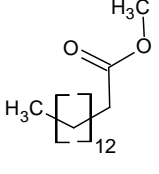
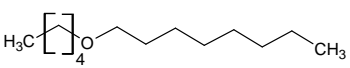
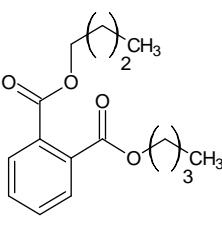
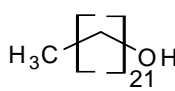
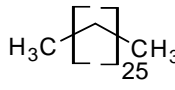
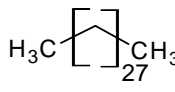
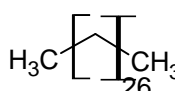
S. No	R. Time	Name of the compound		Molecular formula, molecular weight	MS fragmentions	Uses
1.	4.615	Acetophenone, 4-hydroxy-3, 5-dimethoxy, oxime, bis-TMS		$C_{16}H_{29}NO_4Si_2$	$M^+$ 106, 94, 77, 63, 51, 50	Unknown
2.	23.140	7-Octadecyne		$C_{18}H_{34}$ , 250	$M^+$ 250, 137, 124, 109, 85, 71, 58, 55	Laboratory uses
3.	25.450	Butyl tridecyl ether		$C_{17}H_{36}O$ , 256	$M^+$ 256, 227, 213, 199, 185, 171, 157, 143, 129, 115, 98, 85, 73, 60, 55	Solvent in paintings
4.	26.065	Succinic acid, cyclohexylmethyl 2-pentyl ester		$C_{16}H_{28}O_4$ , 284	$M^+$ 284, 241, 157, 143, 115, 101, 88, 70, 55	Catalyst component for olefin polymerization
5.	34.715	Cyclobutanecarboxamide, N-decyl-N-methyl		$C_{16}H_{31}NO$ , 254	$M^+$ 253, 207, 155, 141, 127, 113, 99, 85, 71, 57, 55	Pharmaceutical uses
6.	35.565	1,2-Benzene dicarboxylic acid, dibutyl ester		$C_{16}H_{22}O_4$ , 278	$M^+$ 279, 167, 149, 113, 104, 71, 57, 55	Additive to adhesives or printing inks.
7.	36.240	Decane		$C_{10}H_{22}$ , 142	$M^+$ 141, 127, 113, 99, 85, 71, 57, 55	Industrial purposes
8.	37.705	Undecane		$C_{11}H_{24}$ , 156	$M^+$ 156, 141, 127, 113, 99, 85, 71, 57, 55	Mild sex attractant
9.	39.120	1-Hexanamine, N-isopropylidene		$C_9H_{19}N$ , 141	$M^+$ 156, 141, 127, 113, 99, 85, 71, 57, 55	In Synthesis, Pharmaceuticals
10.	40.490	9-Octadecenamide		$C_{18}H_{35}NO$ , 281	$M^+$ 281, 207, 141, 127, 113, 99, 85, 71, 57, 55	Slip agent, a lubricant, and a corrosion inhibitor.

 Table Phytochemicals identified in ethanolic extracts of *S. rebaudiana* leaves by GC-MS (RS-4)

S. No.	R. Time	Name of the compound		Molecular formula, molecular weight	MS fragmentions	Uses
1.	4.620	Benzene, 1,3-dimethyl		$C_{10}H_{12}$ , 142	$M^+$ 106, 77, 60, 51	Pain killer
2.	19.145	1-Hexene, 2,4,6-tris-trimethylsilyl		$C_{15}H_{36}Si_3$ , 300	$M^+$ 475, 429, 417, 401, 346, 327, 315, 300	Unknown
3.	23.145	Methyl farnesoate		$C_{16}H_{26}O_2$ , 250	$M^+$ 250, 210, 179, 165, 151, 124, 109, 85, 71, 58, 55	Soaps, cosmetics

# Gc-MS Analysis of Ester And Ethanol Extracts of Stevia Rebaudiana (Leaves)

4.	24.750	Hexadecanoic acid methyl ester		$C_{18}H_{36}O_2$ , 284	$M^+$ 270, 239, 227, 213, 199, 185, 171, 143, 129, 101, 87, 74, 55	Pharmaceuticals
5.	25.475	Pentadecanoic acid methyl ester		$C_{16}H_{32}O_2$ , 256	$M^+$ 256, 227, 213, 199, 185, 171, 157, 143, 129, 115, 98, 85, 73, 60, 55	Pharmaceuticals
6.	26.070	Nonyl pentyl ether		$C_{13}H_{28}O$ , 200	$M^+$ 284, 239, 213, 199, 185, 171, 157, 143, 130, 115, 101, 88, 70, 55	Laboratory uses
7.	35.570	1,2-Benzene dicarboxylic acid, dibutyl ester		$C_{16}H_{22}O_4$ , 278	$M^+$ 279, 168, 167, 149, 132, 113, 104, 84, 71, 57, 55	Pharmaceuticals
8.	36.245	Tricosanol		$C_{22}H_{46}O$ , 326	$M^+$ 366, 356, 343, 327, 315, 295, 282, 253, 225, 211, 207, 195, 169, 156, 141, 127, 113, 99, 71, 57, 55, 53	Bactericidal effect
9.	37.710	Heptacosane		$C_{27}H_{56}$ , 380	$M^+$ 380, 355, 346, 267, 251, 211, 207, 183, 177, 155, 141, 127, 113, 99, 85, 71, 57, 55	Petroleum jelly, cosmetic skin Care
10.	40.490	Nonacosane		$C_{29}H_{60}$ , 408	$M^+$ 408, 355, 282, 249, 225, 207, 183, 169, 155, 141, 127, 113, 99, 85, 71, 57, 55, 54	Petroleum jelly, petrolatum, white petrolatum or soft paraffin
11.	39.125	Octacosane		$C_{28}H_{58}$ , 394	$M^+$ 394, 357, 265, 209, 183, 169, 155, 141, 127, 113, 99, 85, 71, 57, 55	Vaseline, skin lotions and Cosmetics

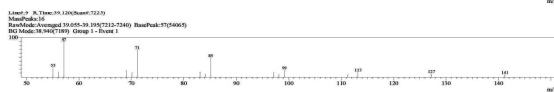
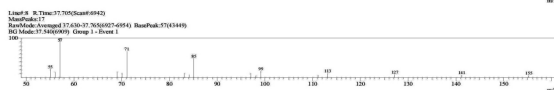
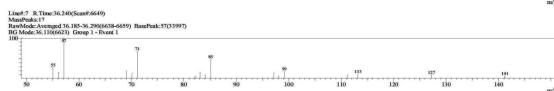
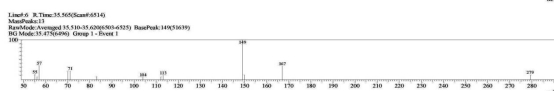
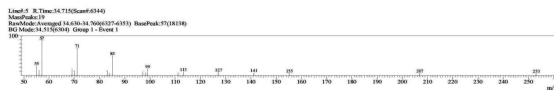
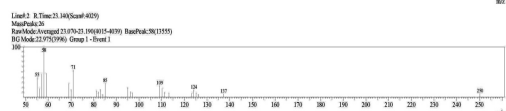
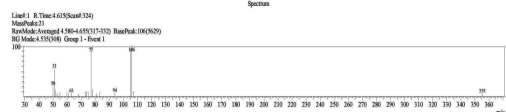
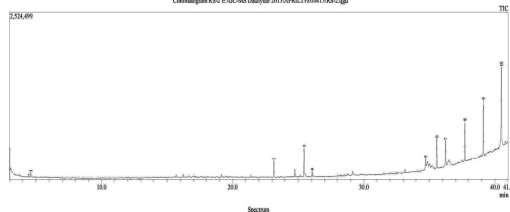
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Sample Information

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## RESULTS AND DISCUSSION

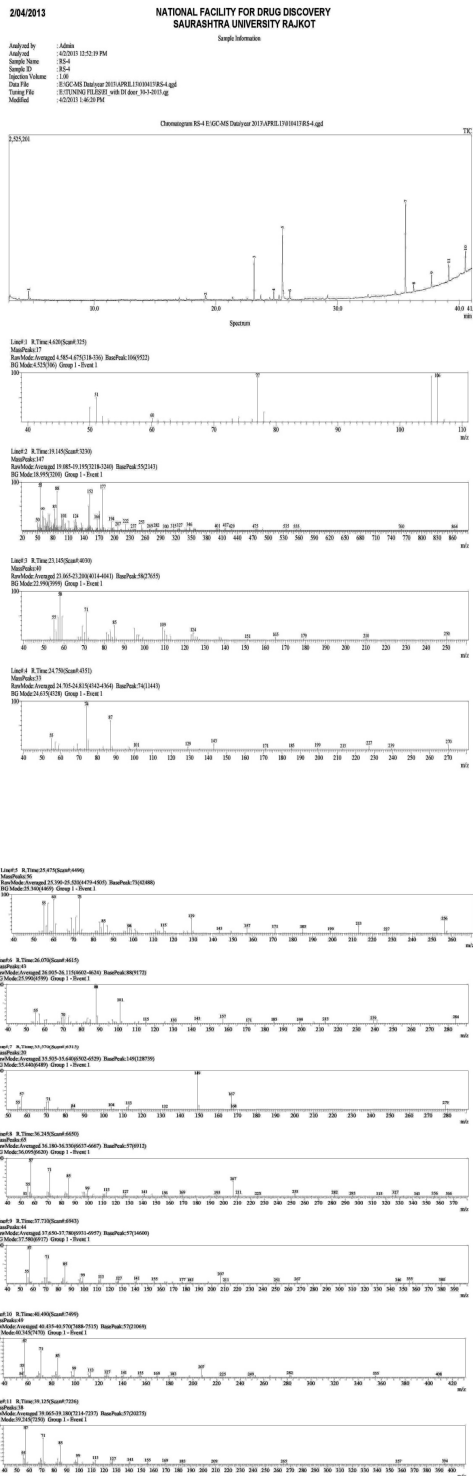
*Stevia* is a model genus for research in ecology and evolutionary biology. The diversity and economical importance of the genus makes it an obvious choice for ecological and evolutionary studies. The objective of the present investigation was to analyze the ester and ethanol extract of the *Stevia rebaudiana* leaves and characterize various phytoconstituents and recognize their industrial and pharmaceutical importance.

### Acknowledgement

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