GC–MS Profiling of Bioactive Constituents in Navachara Kulambu: A Traditional Herbomineral Siddha Medicine

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Abstract

Navachara Kulambu is a traditional Siddha herbomineral preparation used for the management of liver disorders , urinary tract infections, constipation and inflammatory conditions. In the present study, the ethanolic extract of Navachara Kulambu was subjected to Gas Chromatography–Mass Spectrometry (GC–MS) analysis to identify its bioactive phytoconstituents. The chromatogram revealed multiple peaks corresponding to pharmacologically important compounds, including 2-Carboxymethyl-3-methyl-cyclopentane carboxylic acid, Acetic acid, 3,3,6-trimethyl-4-oxo-3,4-dihydro-2H-pyran-2-yl ester, 1-Octene, 2-methoxy, 2-Propenamine, 3-(1-cyclohexenyl)-N-cyclohexyl, N-oxide, and 1,3,12-Nonadecatriene. These compounds are reported to possess antioxidant, anti-inflammatory, antimicrobial, and neuroprotective activities. The results provide a scientific basis for the medicinal use of Navachara Kulambu and support its potential for future pharmacological validation.

Keywords: Navachara Kulambu, Siddha medicine, herbomineral formulation, GC–MS, bioactive phytoconstituents.

Introduction:

Siddha system of medicine, one of the oldest traditional systems practiced in South India, relies on both herbal and herbomineral preparations for therapeutic applications. Navachara Kulambu is a classical Siddha formulation prescribed for liver disorders, inflammatory diseases, and urinary tract infections. Despite its wide medicinal use, there is limited scientific validation of its phytochemical constituents.

Gas Chromatography–Mass Spectrometry (GC–MS) is a powerful analytical technique widely employed to identify volatile and semi-volatile bioactive compounds in complex formulations. By characterizing the phytoconstituents present in Navachara Kulambu, this study aims to provide insight into its pharmacological potential and scientific justification for its traditional therapeutic applications.

Materials and Methods

Preparation of Navachara Kulambu Extract

Ingredients:

• Purified Navacharam (Ammonium chloride)

• Kodikalli paal (latex of Sarcostemma acidum)

Method of preparation :

• The Purified Navacharam is powdered and grind with Kodikalli Paal continuously for three hours. When the mixture attains a thick syrupy consistency, taken store in a container. The extract was prepared using methanol in Soxhlet apparatus and concentrated under reduced pressure.

Drug collection and authentication:

All ingredients of the drug were bought from reputed siddha drug store and authenticated at department of Gunapadam, Govt Siddha Medical College, Palayamkottai.

GC-MS Procedure

Gas chromatography (GC) analysis was carried out using Agilent 6890N gas chromatography equipped with photon multiplier tube as detector coupled to front injector type 1079. The chromatograph was fitted with HP 5 MS capillary column (30 m ×0.25 mm i.d., film thickness 0.25 _m). The injector temperature was set at 250°C, and the oven temperature was initially at 70 °C hold for 4 mins then programmed to 200°C at the rate of 10°C/min and finally held at 200 °C for 13 min. Helium was used as a carrier gas with the flow rate of 1.5 ml/min. 0.2 microlitre of the Drugs PAS/UAS (diluted with methanol 1:10) were injected in the splitless mode. GC–mass spectrometry (GC–MS) analysis of sample was performed using Agilent gas chromatography equipped with JEOL GC MATE-II HR Mass Spectrometer. GC conditions were the same as reported for GC analysis and the same column was used. The mass spectrometer was operated in the electron impact mode at 70 eV. Ion source and transfer line temperature was kept at 250°C. The mass spectra were obtained by centroid scan of the mass range from 50 to 600 amu. The compounds were identified based on the comparison of their retention indices (RI), retention time (RT), mass spectra of WILEY, NIST library data of the GC-MS system and literature data (Adams, 2009).

Derivatization procedure:

For the crude ethanol extracts, a small amount of concentrated sample was taken in a separating funnel and shaken by adding water and ethyl acetate in the ratio of 1:4. The upper layer was collected and concentrated in rotary evaporator to about 1.5 ml. Added 100µl N, O-Bis(trimethylsilyl)trifluoroacetamide and trimethyl chlorosilane (BSTFA+TMCS) and 20µl pyridine and heated at 60°c for 30 minutes. For the layers which are separated from the crude extracts, a small amount of extract was taken and evaporated out totally. To this added acetonitrile and filtered into a conical flask. To the filtrate added 50µl BSTFA+TMCS and heated at 60°c in a water bath for 30 minutes. Filtered using 0.45µ membrane filter to a vial.

Results and Discussions:

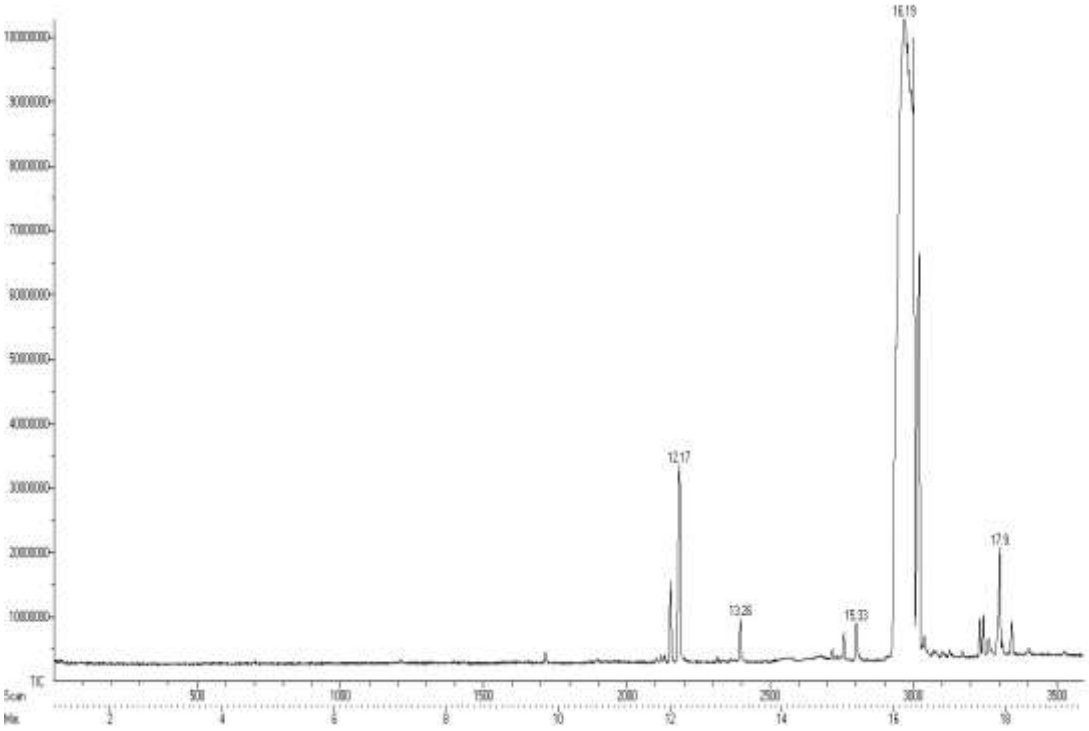
Gas chromatography mass spectroscopy analysis was carried out in crude extracts of the NK such as ethanol extract. The peaks in the chromatogram were integrated and were compared with the database of spectrum of known components stored in the GC-MS library. The detailed of GC-MS analysis of the extracts are given in figures.. Phytochemical analysis by GCMS analysis of the NK revealed the presence of different fatty acids, heterocyclic and phenolic compounds etc.

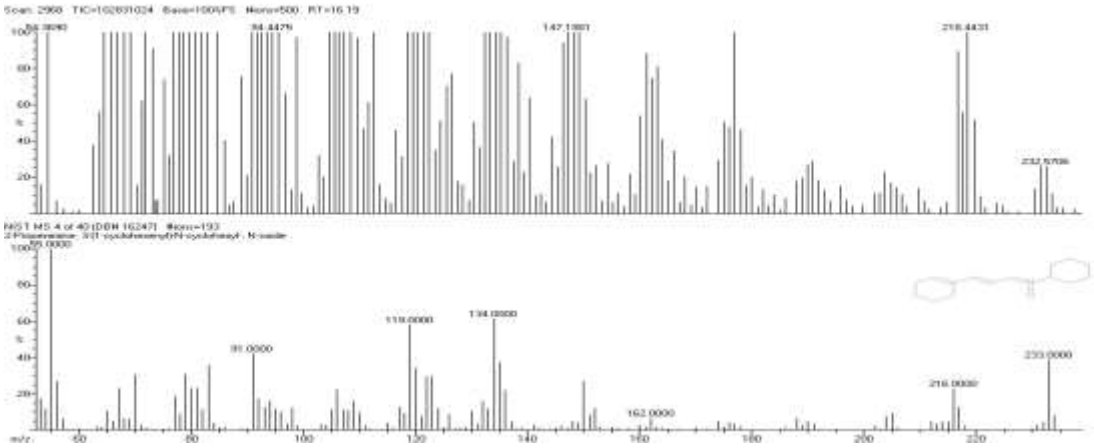
Chromatographic Profile

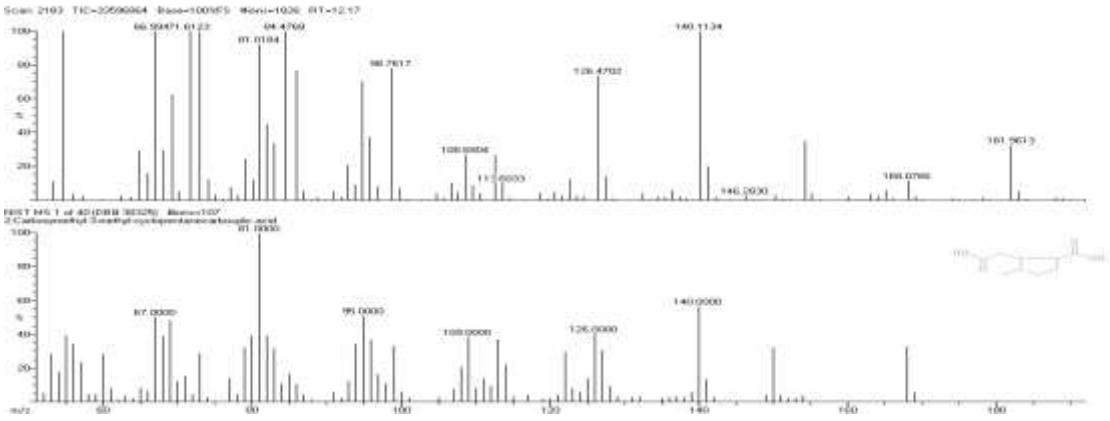
The GC–MS chromatogram of Navachara Kulambu revealed several prominent peaks at retention times 12.17, 13.26, 15.33, 16.19, and 17.9 min, indicating the presence of multiple phytoconstituents.

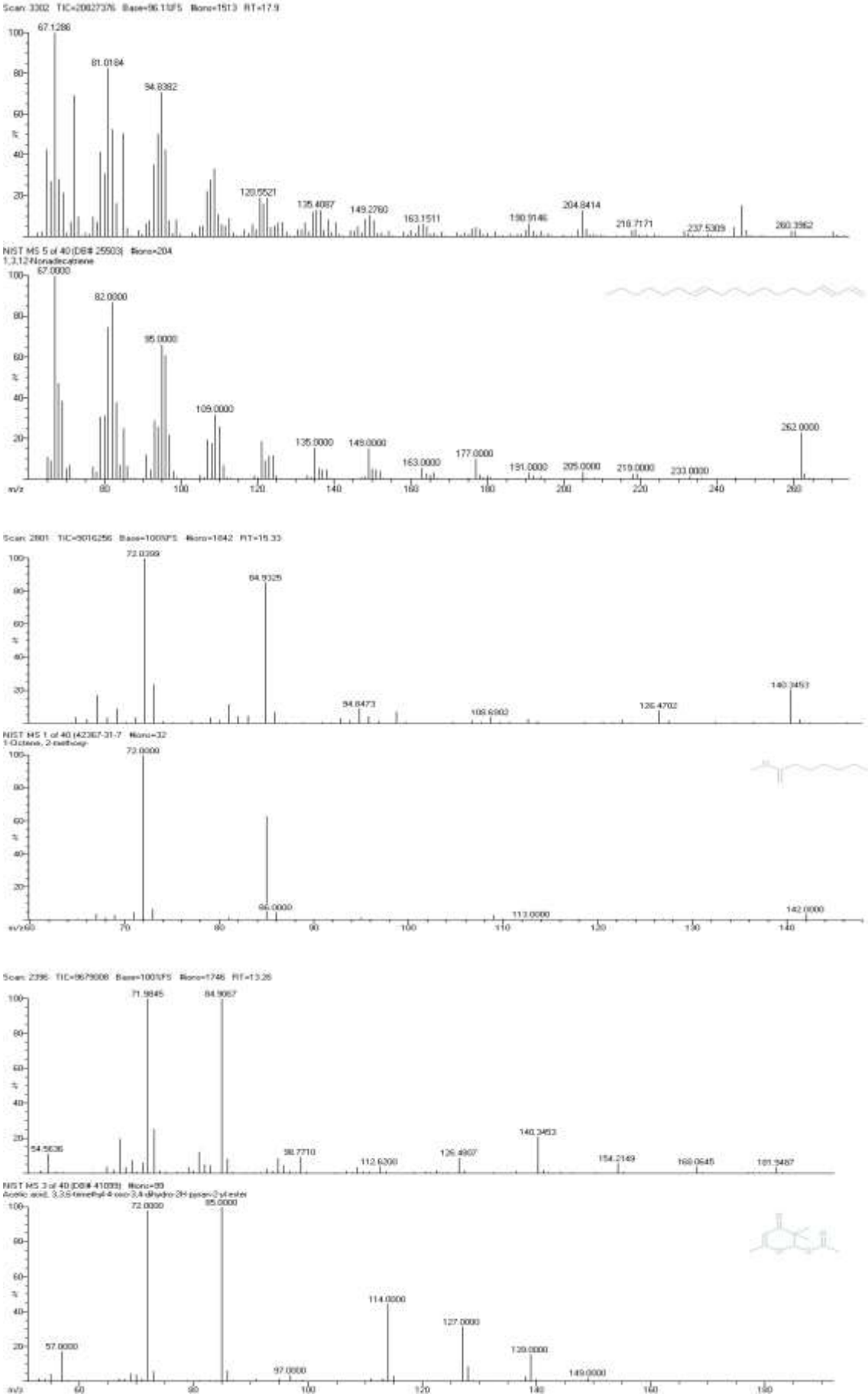
Identified Compounds

Retention Time (min)	Compound Name	Molecular Weight (g/mol)	Reported Biological Activity
12.17	2-Carboxymethyl-3-methyl-cyclopentane carboxylic acid	172	Anti-inflammatory, antioxidant
13.26	Acetic acid, 3,3,6-trimethyl-4-oxo-3,4-dihydro-2H-pyran-2-yl ester	168	Antibacterial, antifungal
15.33	1-Octene, 2-methoxy	142	Antioxidant, antimicrobial
16.19	2-Propenamine, 3-(1-cyclohexenyl)-N-cyclohexyl, N-oxide	233	Neuroactive, antimicrobial
17.90	1,3,12-Nonadecatriene	262	Antioxidant, anticancer, antimicrobial


16.19
12.17
13.26
15.33
17.9
TIC
Scan
Min






67.1286
81.0184
94.8382
120.5521
135.4087
149.2760
163.1511
190.9146
204.8414
218.7171
237.5309
260.3962
1,3,12-Nonadecatriene
67.0000
82.0000
95.0000
109.0000
135.0000
149.0000
163.0000
177.0000
191.0000
205.0000
219.0000
233.0000
262.0000
m/z
72.0269
84.9325
94.8473
109.6902
126.4702
140.3453
1-Octene, 2-methoxy-
72.0000
96.0000
113.0000
142.0000
71.9845
84.9967
54.5636
98.7710
112.6200
126.4907
140.3453
154.2149
168.0645
181.9487
72.0000
85.0000
114.0000
127.0000
57.0000
97.0000
120.0000
149.0000

The GC–MS analysis of Navachara Kulambu demonstrated the presence of bioactive compounds with potential therapeutic effects. Fatty acid derivatives and esters identified are known for their antioxidant and antimicrobial roles, which may contribute to its use in treating inflammatory and infectious conditions. The presence of 1,3,12-Nonadecatriene, with reported anticancer and antimicrobial properties, highlights the pharmacological richness of the formulation. The herbomineral nature of the preparation further suggests possible synergistic effects between phytoconstituents and mineral components, enhancing bioavailability and therapeutic efficacy. These results provide a preliminary chemical profile supporting the Siddha claim of Navachara Kulambu as a potent medicine for ocular and systemic diseases.

Conclusion:

Navachara Kulambu, a traditional Siddha herbomineral preparation, contains multiple bioactive phytoconstituents as confirmed by GC–MS analysis. The GCMS analysis of various compounds from NK extracts was performed using JEOL GC-Mate-II with HP- 5 capillary column and typical total ion chromatograms (TIC) of each sample were given in fig. . The comparison of the mass spectrums with the data base gave more than 90% match as well as confirmatory compound structure match. The GCMS analysis of the concentrated ethanol extract resulted many compounds which have diverse in use. The concentrated ethanol extract contains a variety of fatty acids and derivative compounds. The identified compounds exhibit antioxidant, antimicrobial, and anti-inflammatory activities, justifying its ethnomedicinal use. Further in vitro and in vivo studies are required to validate its pharmacological potential and safety for clinical applications.

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