GC-MS Analysis of an Ayurvedic medicine “Modified Arjunarishta”

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Abstract

Modified Arjunarishta is an ayurvedic formulation used for treatment of cardiovascular diseases. The main ingredients of this formulation are *Terminalia arjuna* bark, *Vitis vinifera* fruit and *Mudhaca longifolia* flower. This is a liquid formulation. GC MS analysis of this formulation is done in the present study to know the various bioactive compounds present therein. It was found that major peaks represented 2,3-dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one, 4-ethyl-2-hydroxycyclopent-2-en-1-one and 1,1-dichloro-2-propanone as compared with NIST software associated instrument. Further work is in progress to prove the validity of this medicine.

Keywords: Arjunarishta, *Terminalia arjuna*, GC MS analysis.

Introduction

Ayurveda is an oldest serving complete medical system in the world. The word Ayurveda derived from ancient Sanskrit in which ‘Ayus’ means life and ‘ved’ means knowledge. It originates back to 5000 years. There are spread around sixth century BC to Tibet, China, Mongolia, Korea, and Sri Lanka carried over by Buddhist monks travelling to these lands. The prime raw material of Ayurveda are the medicinal plants. Around 8,000 species of medicinal plants are being used in India for betterment of health (Anon, 1996). Some of the factors for this state of Ayurveda and Siddha medical system is discussed by Raj et al, 2011; Rao et al, 2015a, b; Ravi et al, 2015). Pordie’ and Gaudillie’re 2014, have discussed in details about the factors involved in polyherbal formulation in drug discovery and related issues pertaining to Ayurveda as an industry.

Arishta and Asava have been used as medicines for over 3000 years to treat various disorders and also taken as stimulants and appetizers. In Ayurveda, Arishtas are an important formulations. Arjunarista also said as parthadayarishta is one of the ancient liquid oral formulations prescribed in Ayurveda for cardiovascular disorders. It nourishes and strengthens cardiac muscle and there function by regulating cholesterol and blood pressure. Usually there ingredients are *Terminalia arjuna*, *Madhuca indica*, *Vitis vinifera*, *Woodfordia fruticosa* and Jaggery. As it is a formulated product, a modulatory form of Arjunarista is used to bring alcohol free. The studying of Modified Arjunarista’s ingredients are *Terminalia arjuna*, *Madhuca longifolia* and *Vitis vinifera* (Raisins).

The formulation was prepared by making a decoction of these three plants in specified amounts as listed in AFI.

A knowledge of the phytochemical constituents of plants is desirable not only for the discovery of therapeutic agents, but also because such information may be of great value in disclosing new sources of economic...
phytocompounds for the synthesis of complex chemical substances and for discovering the actual significance of folkloric remedies (Milne et al., 1993). Hence a thorough validation of the Ayurvedic formulation has emerged as a new branch of science emphasizing and prioritizing the standardization of the natural drugs and products because several of the phytochemicals have complementary and overlapping mechanism of action. Mass spectrometry, coupled with chromatographic separations such as Gas chromatography (GC/MS) is normally used for direct analysis of components existing in traditional medicines and medicinal plants. In recent years GC-MS studies have been predominantly applied for the analysis of medicinal plants as this technique has proved to be a valuable method for the analysis of non polar components and volatile essential oil, fatty acids, lipids (Jie et al., 1991) and alkaloids (Betz et al., 1997). Chromatography is the term that describe a separation technique in which a mobile phase carrying a mixture is caused to move in contact with a selectively absorbent stationary phase. It also plays a basic role as an analytical technique for quality control and standardization of phyto therapeutics (Andrew, 2007). In the analysis of medical plant extracts especially ayurvedic medicine, there was a minimum of two significant advantages for GC-MS. These include; (a) Using the capillary column, GC-MS has a fine seperation ability that capable of producing chemical finger print of high quality, (b) Using the coupled mass spectroscopy and the corresponding mass spectral data base, the qualitative and relatively quantative compositonn information of the ayurveda arjunarishta investigatd could be provided by GC-MS which is extremely useful for future research for eluciding the relationship between chemical constituents in the herbal medicine and there pharmacology in future research. In this way it is suggested that GC-MS is mostly preferable tool for the analysis of volatile chemical constituents in medical plants.

The aim of the present study is to identify the phyto compounds of Modified Arjunarishta using GC-MS. The work was enable to identify the phytochemicals that responsible for cardiotherapeutic activity.

Materials and Methods

Collection and Authentication

The selected plants for the proposed study include Terminalia arjuna bark, Vitis vinifera fruit, and Mudhuca longifolia flower which are collected from Villivakkam,chennai, K.G. Kandigai, tiruttani and from Ayurvedic shop. These are identified and authenticated by Mr. K.N. Sunilkumar, Research officed (Pharmacognosy) from Siddha central research institute, Chennai.

Preparation of the Modified Arjunarishta

The identified plant powers of Terminalia arjuna bark, Vitis vinifera fruit, and Mudhuca longifolia flower were mixed with 32 part of sterile water and allow to boil. The boiling was continued still the liquid level reduced to one part through evaporation. The extract was filtered through filter paper. The filtrate was transferred in to an air tight sterile container and it is ready to use for further analysis.

Gas chromatography was carried out at VIT Vellore. A huge attainable separation power in combination with wide range of detectors employing different detection principles to which it can be coupled makes GC an important tool in the analysis at minute level of plant phytochemical compounds.

Sample preparation

The fluid alcohol free arjunarista is filtered through whatmann filter paper no.41 to remove sediments and precipitates. Prior to filtering, the filter paper was made wet with 95% ethanol. The filtrate was later concentrated by bubbling nitrogen gas in to the solution. 1 ul of this solution was employed for to run GC-MS.

Instrumentation

The phyto compounds analysing GC-MS was GC clarus 680 Parkin elmer make. A fused silica column, packed with Elite – 5ms (5% biphenyl 95% dimethyl polysiloxane) of which dimension 30m x 0.25mm ID x 250um df is used. The compounds were seperated using Helium as career gas at a constant flow of 1ml/min. During the chromatography run, the injector temperature was set at 260°C. Sample volume of 1ul injected into the instrument and the oven temperature was as follows : 60°C for two minutes followed by 300°C, it was hold for six minutes. The mass detector condition were: transfer line temperature 240°C, ion source temperature 240°C and ionisation mode electron impact at 70 eV, a scan time 0.2 sec and the scan interval of 0.1 sec . The fragment ranges from 40 to 600 dalton are analysed. The spectrums of the compounds obtained were compared with the database of spectrum of known compounds stored in the GC-MS NIST 2008 Library. Total run time took 32 minutes.

Results and Discussion

The GC-MS Analysis of modified Arjunarishta were analysed and the results were tabulated in Table 1. The GC-MS Analysis chromatogram of the modified Arjunarishta obtained from tri herbal constituents in shown in figure 1.
From the GC-MS chromatogram many peaks are obtained. But only eight compounds were identified with the comparison of NIST Software available in the instrument and also peak area percentage. It is found that the compound 2,3-dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one was present higher in the formulation (45.329 area %). It shows the more synergic effect of the formulation. Following the compound, 4-ethyl-2-hydroxycyclopent-2-en-1-one was present in moderate concentration in the formulation (32.146 area %). The other compounds present in minimum concentration such as 1,1-dichloro-2-propanone, 2-amino-octadec-7-ene-1,3-diol butaneboronate, Acetamidoacetaldehyde, 2-hydroxy-2-cyclopenten-1-one, Trimethyl[4-(2-methyl-4-oxo-2-pentyl)phenoxy]silane, Hexamethyl-cyclotrisiloxane.

| S.No. | Retention time | Compound name                                           | Molecular weight | Peak area % |
|-------|----------------|----------------------------------------------------------|------------------|-------------|
| 1     | 2.558          | Acetamidoacetaldehyde                                     | 101              | 3.053       |
| 2     | 2.774          | 1,1-dichloro-2-propanone                                  | 127              | 8.489       |
| 3     | 8.126          | 2-hydroxy-2-cyclopent-1-one                               | 98               | 2.795       |
| 4     | 11.212         | 2,3-dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one        | 144              | 45.329      |
| 5     | 11.702         | 2-amino-octadec-7-ene-1,3-diol butaneboronate             | 365              | 3.431       |
| 6     | 13.913         | 4-ethyl-2-hydroxycyclopent-2-en-1-one                     | 126              | 32.146      |
| 7     | 26.868         | Hexamethyl-cyclotrisiloxane                               | 222              | 1.964       |
| 8     | 27.023         | Trimethyl[4-(2-methyl-4-oxo-2-pentyl)phenoxy]silane       | 264              | 2.793       |
2-HYDROXY-2-CYCLOPENTEN-1-ONE

2,3-DIHYDRO-3,5-DIHYDROXY-6-METHYL-4H-PYRAN-4-ONE

2-AMINO-OCTADEC-7-ENE-1,3-DIOL BUTANEBORONATE

4-ETHYL-2-HYDROXYCYCLOPENT-2-EN-1-ONE
Conclusion

The results of GC-MS analysis revealed that the Modified Arjunarishta contains the enriched compounds like 2,3-dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one and 4-ethyl-2-hydroxycyclopent-2-en-1-one. Further investigation on the cardioprotective and therapeutic activity of this Ayurvedic formulation in an in-vivo model is under way.

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