Gas Chromatography-Mass Spectrometry Analysis of Ethanolic Fruit Extract of *Momordica charantia* var. *Muricata*

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**Abstract**

The fruits of *Momordica charantia* (bitter gourd) have been used as folklore medicine for the management of ailments such as leprosy, menstrual problems, hypertension and most efficiently for diabetes. The present investigation was carried to identify active components in small round variety of immature fruit *M. chrantia muricata* was analysed. The fruits were procured directly from the garden at Bhuvanagiri, Tamil Nadu, were washed in tap water and cut into small pieces and were shade dried for 10 days. The dried fruits were ground into fine powder. The sample was extract with ethanol and analyzed through Gas Chromatography-Mass spectrometry for identification of different compounds. Analysis revealed presence of 25 phyto components. The major compound present in the ethanolic fruit extract was identified as β-Sitosterol with RT 37.26 and 12.16% relative peak area. This compound has antihyperlipoproteinaemic, antibacterial and antimicotic activity, and helps tumor inhibition in vivo.

**Keywords:** GC-MS Analysis, *Momordica charantia* var. *muricata*, Phyto Components

1. Introduction

“*Momordica charantia* L. commonly known as bitter gourd is an economically important medicinal plant belonging to the family cucurbitaceae. Two varieties of this plant are cultivated in India. *M. charantia* var. *charatia* with large fruits are fusiform in shape and *M. charantia* var. *muricata*, are identified by small, round fruit”. The small round variety of fruits are 2-3 cm long and 1 cm diameter. “All parts of plant, especially roots, leaves, fruits and seeds are widely used as traditional medicine throughout Asia, East Africa and South America. The leaves and fruits are used for external application in lumbago, ulceration and bone fracture and internally in leprosy, haemorrhoids and Jaundice”. “Fruits and seeds of bitter gourd possess medicinal properties such as anti-HIV, anti-ulcer, anti-inflammatory, anti-leukemic, anti-microbial, anti-tumor and antidiabetic property”. “The immature fruits are eaten as vegetables and are good sources of vitamin C, vitamin A and phosphorus and iron”. Review of literature did not reveal much evidence on the phyto components available in small variety of bitter gourd; hence the present research was done to find out the active constituents present in the fruit extract.

2. Materials and Methods

2.1 Plant Material

The availability of bitter gourd was identified in plenty from the gardens in the Bhuvanagiri village, Cuddalore district, Tamil Nadu and the variety of small bitter gourd was authenticated by the Department of Pharmagognosy,
2.2 Preparation of Plant Material

Fresh unripe fruit of *Momordica charantia* var. *muricata* was procured from the garden and was washed in running water to remove dust and any other foreign materials, sliced into round and very thin pieces and was dried in shade until the unripe fruit pieces were void of moisture and brittle. The dried pieces were ground into a powder with domestic electric grinder.

2.3 Gas Chromatography and Mass Spectrometry Analysis

Twenty five gram of sample was soaked overnight in 60ml ethanol. After filtration 1ml of filtrate was evaporated to 2µl. The components were analyzed using Bruker 436GC column and BR-5MS with a heating rate of

| No. | RT   | Name of the compound                                   | Molecular Formulae | Molecular Weight | Peak Area % |
|-----|------|--------------------------------------------------------|--------------------|------------------|-------------|
| 1   | 4.69 | 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydoxy-6-methyl-     | C₆H₈O₄             | 144              | 0.4         |
| 2   | 7.89 | Eugenol                                                | C₁₀H₁₂O₂            | 164              | 2.52        |
| 3   | 11.03| 6-Epishyobunone                                        | C₁₅H₂₄O₂            | 220              | 0.12        |
| 4   | 13.48| 5,5,8a-Trimethyl-3,5,6,7,8,8a-hexahydro-2H-chromene    | C₁₂H₂₀O             | 180              | 0.71        |
| 5   | 15.07| Hexadecanoic acid, methyl ester                        | C₁₇H₃₄O₂            | 270              | 0.63        |
| 6   | 15.99| Hexadecanoic acid, ethyl ester                         | C₁₈H₃₆O₂            | 284              | 2.24        |
| 7   | 17.39| 9,12-Octadecadienoic acid (Z,Z)-                       | C₁₈H₃₂O₂            | 280              | 0.46        |
| 8   | 17.64| Phytol                                                 | C₁₉H₃₂O₂            | 296              | 0.72        |
| 9   | 18.34| Linoleic acid ethyl ester                              | C₂₀H₄₀O₂            | 308              | 0.61        |
| 10  | 18.83| Octadecanoic acid, ethyl ester                         | C₂₀H₄₀O₂            | 312              | 4.52        |
| 11  | 20.53| 9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z,Z)-   | C₂₀H₃₄O₂            | 306              | 1.94        |
| 12  | 21.13| Eicosanoic acid                                        | C₂₀H₄₀O₂            | 312              | 1.14        |
| 13  | 22.88| 5,8,11,14-Eicosatetraenoic acid, methyl ester, (all-Z)- | C₂₁H₄₁O₂            | 318              | 5.88        |
| 14  | 23.53| Glycidol stearate                                      | C₂₁H₄₀O₄            | 340              | 0.05        |
| 15  | 25.05| Butyl 9,12,15-octadecatrienoate                        | C₂₂H₄₃O₂            | 334              | 16.36       |
| 16  | 25.71| 5,8,11,14-Eicosatetraenoic acid, ethyl ester, (all-Z)- | C₂₂H₄₃O₂            | 332              | 5.8         |
| 17  | 26.49| Octadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester| C₂₁H₄₁O₄            | 358              | 9.14        |
| 18  | 27.73| Squalene                                               | C₃₀H₄₀               | 410              | 4.89        |
| 19  | 28.11| 9,12,15-Octadecatrienoic acid, 2,3-bis{[trimethylsilyl]oxy}propyl ester, (Z,Z,Z)- | C₃₄Hₕ₂O₈Si₂         | 496              | 2.95        |
| 20  | 31.3 | γ-Tocopherol                                           | C₂₉H₄₈O₂            | 416              | 3.15        |
| 21  | 32.81| Vitamin E                                              | C₂₉H₄₈O₂            | 430              | 5.69        |
| 22  | 36.77| Stigmasterol                                            | C₂₉H₄₈O₂            | 412              | 7.65        |
| 23  | 37.26| β-Sitosterol                                            | C₂₉H₄₈O₂            | 414              | 12.16       |
| 24  | 38.47| Stigmasta-5,24(28)-dien-3-ol, (3β)-                   | C₂₉H₄₈O₂            | 412              | 8.67        |
| 25  | 38.96| Ergost-7-en-3-ol, (3β)-                               | C₂₉H₄₈O₂            | 400              | 1.6         |

RT – Retention Time
10°C/minute, sample injection was performed in the split ratio 10:1 and the carrier gas flow was maintained at 1 ml/minute during the run.

### 3. Results and Discussion

The analysis of the ethanolic extract of bitter gourd powder of *M. charantia* indicates 25 different compounds at different Retention Time (RT) and different percentage peak area are listed in Table 1. Interpretation of GC-MS was conducted using the database of NIST Version 11. The comparison of the mass spectrums with the data base gave more than 90% match as well as confirmatory compound of the structure match. The peak compounds were compared with the known phyto-constituents and the molecular weight, structure and formula were ascertained by using the NIST, Pub Chem, Chem Spider and Chemical book.

The Table 1 shows the different compounds identified in the ethanolic extract of bitter gourd powder with different retention time and peak area.

The major compound present in *M. charantia* was β-Sitosterol with RT 37.26 and 12.16 % relative peak area. The structure of β-Sitosterol is given in Figure 1, the molecular formula is **C_{29}H_{50}O**, the molar weight is 414 and it has anti-hyperlipoproteinaemic, antibacterial, antimicotic activity and is a inhibitor of tumor promotion.

#### 3.1 Therapeutic Properties of Phyto Compounds

The therapeutic properties of phytocompounds present in the ethanolic extract of small round bitter gourd powder of *Momordica charantia* var. *muricata* are shown in the Table 2.

1. Antifungal 2. Antimicrobial 3. Antioxidant 4. Anti-inflammatory 5. Cardiovascular properties 6. Hypocholesterolemic 7. Nematicide 8. Pesticide 9. Diuretic

### Table 2. Phyto compounds and functional properties in unripe fruit powder of *M. charantia*

| Compounds | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 29 |
|-----------|---|---|---|---|---|---|---|---|---|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl- | + | + |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Eugenol   | + | + | + |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Hexadecanoic acid, methyl ester | + | + | + |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Hexadecanoic acid, ethyl ester | + | + | + |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Phytol    | + | + | + |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| 9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z,Z)- | + | + | + |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Octadecanoic acid, ethyl ester | + |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Squalene  | + | + |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Vitamin E | + | + | + |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Stigmasterol | + |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| β-Sitosterol | + | + |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Stigmasterol, 5,24(28)-dien-3-ol, (3β)- | + |   |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
| Ergost-7-en-3-ol, (3β)- | + | + |   |   |   |   |   |   |   |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |    |
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**4. Conclusion**

In GC-MS analysis, 25 active components were identified in ethanolic extract of small round variety of *M. charantia*. These active compounds help to reduce the inflammation, has hyperglycemic effect, polyurea, hypercholesterlimia, leukemia, cancer, cardiovascular disease and ulcer. They also eliminate toxic substance present in various parts of the body and thus show a positive effect on health.

**5. References**

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**Figure 1.** β-Sitosterol.