Frontiers in Health InformaticsISSN-Online: 2676-7104 www.healthinformaticsjournal.com
2025; Vol 14: Issue 2 Open Access

PHYTOCHEMICAL PROFILING OF MOMORDICA CHARANTIA L., SEEDS USING GC-MS ANALYSIS

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Cite this paper as: SUBITHA SHAJINI R, ABISHA K (2024), PHYTOCHEMICAL PROFILING OF MOMORDICA CHARANTIA L., SEEDS USING GC-MS ANALYSIS. *Frontiers in Health Informatics*, *14(2) 2448-2460*

Abstract: This research focuses on the phytochemical profiling of *Momordica charantia* seeds using Gas Chromatography-Mass Spectrometry (GC-MS) analysis. *Momordica charantia*, also known as bitter melon, has been widely used in traditional medicine for its various biological properties, including anti-diabetic, anti-inflammatory, and antioxidant activities. The general objective of this study is to explore the ethnobotanical uses, biological properties, and chemical composition of *Momordica charantia* seeds. The study is conducted in two phases: Phase I involves the collection of *Momordica charantia* seeds, followed by extraction using aqueous and methanol solvents. Preliminary phytochemical tests are performed to identify bioactive compounds such as alkaloids, flavonoids, and saponins. In Phase II, the extracts are subjected to GC-MS analysis to identify and characterize the isolated compounds. The GC-MS technique will provide a detailed chemical profile of the seeds, offering insights into the specific compounds responsible for their medicinal properties. This study aims to contribute to a deeper understanding of the pharmacological potential of *Momordica charantia* seeds, supporting their traditional uses while uncovering new therapeutic applications.

Keywords: *Momordica charantia* L., GC-MS Analysis, phytochemical profiling, medicinal properties, bioactive compounds, ethnobotany.

INTRODUCTION

Momordica charantia L., commonly known as bitter melon, is a tropical and subtropical plant belonging to the Cucurbitaceae family. This plant, widely distributed in Asia, Africa, and the Caribbean, has a long history of use in traditional medicine, particularly for managing conditions like diabetes, gastrointestinal disorders, and infections (Ravikumar *et al*., 2020). Its fruit, leaves, and seeds are known for their broad spectrum of medicinal properties, but the seeds have garnered attention for their unique phytochemical composition and therapeutic potential. The bitter taste of the plant's fruit, a characteristic feature of *Momordica charantia*, is due to compounds like charantin, which contribute to its anti-diabetic effects (Khan *et al*., 2019). However, the seed's bioactive constituents remain less studied, and their exact chemical composition is not fully understood. Recent research has highlighted the promising pharmacological activities of *Momordica charantia* seeds, including anti-inflammatory, antioxidant, and antimicrobial properties (Baskaran *et al*., 2020). These properties are attributed to the various bioactive compounds present in the seeds, such as alkaloids, flavonoids, saponins, terpenoids, and phenolic acids (Ravikumar *et al*., 2020). These compounds are known to interact with different metabolic pathways and cellular processes, thus influencing various disease mechanisms. For instance, *Momordica charantia* seeds have shown significant promise in managing metabolic disorders, particularly Type

2 diabetes, by improving insulin sensitivity and reducing blood glucose levels (Akram *et al*., 2019). Moreover, recent studies have indicated that compounds in *Momordica charantia* seeds exhibit potent anti-cancer, anti-obesity, and neuroprotective effects, further increasing interest in this plant's medicinal potential (Liu *et al*., 2021).

Despite the therapeutic potential of *Momordica charantia* seeds, there is a lack of comprehensive studies profiling their chemical constituents. Previous studies have primarily focused on the fruit and leaves, leaving the seeds underexplored in terms of their phytochemical and pharmacological properties. Gas Chromatography-Mass Spectrometry (GC-MS) analysis, a widely used and effective technique for profiling plant metabolites, offers a detailed understanding of the chemical composition of plant extracts. By applying this technique to *Momordica charantia* seeds, it is possible to isolate and identify a variety of compounds that contribute to their medicinal effects, thus enhancing the scientific knowledge of the plant's full pharmacological potential.

The general objective of this study is to conduct a comprehensive review of the ethnobotany, uses, and biological properties of *Momordica charantia* seeds, followed by an analysis of their chemical constituents through GC-MS. The study is divided into two phases: Phase I focuses on the collection, extraction, and preliminary phytochemical testing of the seeds, while Phase II involves the GC-MS analysis of the extracts to identify and characterize the bioactive compounds. This study aims to bridge the knowledge gap by providing a detailed chemical profile of *Momordica charantia* seeds and contributing to the understanding of their potential uses in medicinal applications.

As the world continues to seek alternative and complementary therapies for a variety of diseases, plants like *Momordica charantia* offer significant promise. This research not only aims to characterize the chemical makeup of *Momordica charantia* seeds but also to explore its potential therapeutic benefits, which may lead to the development of novel natural remedies for the management of various health conditions.

MATERIALS AND METHODS

MATERIALS

Momordica charantia, commonly known as bitter gourd, bitter melon, or karela, is a member of the Cucurbitaceae family. It is native to humid and subtropical regions worldwide (Anjum *et al*., 2013).

Collection of Plant Material

Fresh seeds of *Momordica charantia* were collected for the study (Fig. 4). The seeds were washed thoroughly and air-dried in the shade at room temperature (Fig. 5). Sun drying, a cost-effective method for drying plant samples, was employed for this purpose. This method requires a minimum temperature of 30 ˚C or higher, and the drying process typically spans several days to ensure the removal of excess moisture from the samples (Banu & Cathrine, 2015). After drying, the samples were stored in dry containers under low humidity conditions to prevent moisture absorption, which could affect the quality of the dried material. The dried seeds were then ground into a fine powder using a mechanical blender or a mortar and pestle (Fig. 6). This powdered material was subsequently used for the extraction process, where the dried sample and solvent were combined in a vessel to initiate the extraction of bioactive compounds.

METHODS

Preparation of Crude Extract

To prepare the crude extract, 50 ml each of methanol and water (aqueous) were measured and added to sterilized 100 ml conical flasks. To this, 5 grams of dried powdered *Momordica charantia* seeds were added. The mixture was soaked for 72 hours to allow the extraction of bioactive compounds. After the soaking period, the extracts were filtered through cheesecloth to separate the solid residues. The supernatants were then

collected, labelled, and stored for subsequent screening of various phytochemicals (Harborne, J. B., 1998) (Fig. 7, 8).

I. Phytochemical Screening for Different Compounds

Detection of Carbohydrates

Benedict's Test: To 0.5 mL of filtrate, 0.5 mL of Benedict's reagent was added. The mixture was heated in a boiling water bath for 2 minutes. A characteristic-colored precipitate indicated the presence of sugar (Raaman, 2006).

Fehling's Test: 1 mL of filtrate was boiled with 1 mL each of Fehling's solutions A and B. A red precipitate indicated the presence of sugar (Raaman, 2006).

Fig1.*Momordica charantia* (plant).

Fig 2. *Momordica charantia* (unripe fruit)

Fig 3.*M. charantia* (ripen fruit)

Fig 4. *M. charantia* (ripen seeds)

Fig.5 *M. charantia* (dry seeds)

Fig 6.*M. charantia* (seed powder)

Test for Cardiac Glycosides

Keller-Killani Test: To 1 mL of test solution, 1.5 mL of glacial acetic acid and 1 drop of 5% ferric chloride were added. Few drops of concentrated sulfuric acid were carefully added along the side of the test tube. Formation of a blue color in the acetic acid layer indicated the presence of cardiac glycosides (Singh & Kumar,

2017).

Detection of Proteins and Amino Acids

Xanthoproteic Test: The extract was treated with a few drops of concentrated nitric acid. The formation of a yellow color indicated the presence of proteins (Tiwari *et al*., 2011).

Fig.7 Methanol extract of *M. charantia* seed ,Fig. 8 Aqueous extract of *M.charantia* seed

Detection of Flavonoids

Ferric Chloride Test: When an aqueous extract was mixed with 10% ferric chloride ($FeCl_3$), a green precipitate formed, indicating the presence of flavonoid compounds (Sani *et al*., 2017).

Sulfuric Acid Test: A fraction of the extract was treated with concentrated H_2SO_4, and the formation of an orange color was observed, indicating the presence of flavonoids (Tyagi, 2017).

Detection of Phenols

Test for Carotenoids: 1 g of filtrate was extracted with 10 mL of chloroform in a test tube with vigorous shaking. The mixture was filtered, and 85% sulfuric acid was added. A blue color at the interface indicated the presence of carotenoids (Tyagi, 2017).

Potassium Dichromate Test: The addition of a few drops of potassium dichromate solution to the plant extract resulted in a color change to dark, indicating the presence of phenols (Kumar *et al*., 2018).

Detection of Phytosterols

Libermann-Burchard's Test: The extract (50 mg) was dissolved in 2 mL acetic anhydride. One or two drops of concentrated sulfuric acid were added slowly along the side of the test tube. The appearance of color changes indicated the presence of phytosterols (Raaman, 2006).

Salkowski's Test: 1 mL of extract was treated with 1 mL of chloroform and filtered. The filtrate was added to a few drops of concentrated sulfuric acid, shaken, and allowed to stand. If the lower layer turns red, a steroid is present, and the golden yellow layer at the bottom indicates triterpenoids (Singh & Ramesh, 2017).

Detection of Terpenoids

2.0 mL of chloroform was added to 5 mL of the aqueous plant extract and evaporated on a water bath. The residue was then boiled with 3 mL of concentrated H_2SO_4. A grey color formed, indicating the presence of terpenoids (Gul *et al*., 2017).

0.5 mL of crude extract, 2 mL of chloroform, and 3 mL of sulfuric acid were mixed, and the formation of a reddish-brown color indicated the presence of terpenoids (Dubale *et al*., 2023).

Detection of Quinones

Sulfuric Acid Test: One drop of concentrated sulfuric acid was added to 10 mg of each extract dissolved in isopropyl alcohol. The formation of a red color indicated the presence of quinones (Maria, 2018).

Detection of Anthraquinones

Ammonium Hydroxide Test: One drop of concentrated ammonium hydroxide was added to 10 mg of each extract, previously dissolved in isopropyl alcohol. After two minutes, the formation of a red color indicated the presence of anthraquinones (Maria *et al*., 2018).

Detection of Coumarins

10% sodium hydroxide solution was added to the extract. If the solution turned yellow, the extract contained coumarins (Kumar *et al*., 2018).

Detection of Emodins

2 mL of NH_4OH and 3 mL of benzene were added to the extract. The appearance of a red color indicated the presence of emodins (Rauf *et al*., 2013).

Detection of Saponins

Sodium Bicarbonate Test: To the plant extract, a few mL of sodium bicarbonate solution were added. After adding distilled water and shaking vigorously, the appearance of stable, honeycomb-like froth indicated the presence of saponins (Ray *et al*., 2013).

Detection of Fixed Oil and Fat

Saponification Test: To small quantities of various test solutions, a few drops of 0.5N alcoholic potassium hydroxide and a drop of phenolphthalein were added. The mixture was heated on a water bath for 1-2 hours. The presence of fixed oils and fats was indicated by the formation of soap (Nanna *et al*., 2013).

Detection of Tannin

The extract was treated with a few drops of lead acetate solution, and the formation of a yellow color solution indicated the presence of tannins (Supraja *et al*., 2015).

Detection of Alkaloids

$FeCl_3$ Test: One drop of $FeCl_3$ solution was added to each of the test samples, and the formation of a yellow precipitate indicated the presence of alkaloids (Archana *et al*., 2012).

Detection of Glycosides

Concentrated Sulfuric Acid Test: Concentrated H_2SO_4 was added to the test sample, and the appearance of a reddish color indicated the presence of glycosides (Brindha *et al*., 1977).

II. Gas Chromatography–Mass Spectrometry Analysis

The GC-MS analysis of volatile compounds in *Momordica charantia* seeds (methanolic extract) was carried out using an Agilent 8890 system, which included an AOC-20i auto-sampler and a Gas Chromatograph connected to a Mass Spectrometer (GC-MS). The system was equipped with an Elite-5MS capillary column (30 × 0.25 µm ID × 0.25 µm film thickness), composed of 5% diphenyl and 95% dimethyl polysiloxane, ideal for separating volatile compounds. For GC-MS analysis, an electron ionization system was operated in electron impact mode with an ionization energy of 70 eV. Helium gas (99.99%) was used as the carrier gas at a constant flow rate of 1.2 mL/min, and the injection volume was 1 µL with a split ratio of 15:1. The injector temperature was set to 250°C, while the ion-source temperature was maintained at 230°C. The oven temperature was programmed to start at 35°C, ramping at a rate of 5°C/min until reaching 180°C, where it was held for 3 minutes. The temperature then increased at 5°C/min to 300°C, with a final hold for 5 minutes. Mass spectra were recorded at 70 eV, with a scan interval of 0.5 seconds, capturing fragments in the mass range of 45 to 450 Da. The solvent

delay was set to 3 minutes, and the total GC-MS run time was 53.5 minutes. The relative quantity of each compound was determined by comparing the average peak area of each compound to the total area. The mass detector used for this analysis was a Turbo-Mass Gold-Perkin Elmer, and the data were analyzed using the Turbo-Mass software version 5.2.28-29 (Prabahar *et al*., 2022).

RESULTS

The phytochemical screening of *Momordica charantia* seed extracts was conducted using aqueous and methanol solvents, and the results of various tests for bioactive compounds are summarized in the Table 1.

Phytochemical Findings

The phytochemical screening of *Momordica charantia* seeds revealed the presence of several bioactive compounds that contribute to the medicinal properties of the plant. Alkaloids, a group of naturally occurring organic compounds with pharmacological activity,

Table 1. Preliminary phytochemical analysis of the aqueous and Methanol extract of *Momordica charantia* L.

Phytochemicals	Test	Aqueous	Methanol
Alkaloids	$FeCl_3$ test	++	+++
Anthraquinones	Ammonium hydroxide test	-	-
Carbohydrates	Benedict's test	-	-
	Fehling's test	-	-
Cardiac Glycosides	Keller-Killani test	-	-
Coumarins	10% NaOH test	+	+
Emodins	NH_4OH test	-	-
Fixed Oil and Fat	Saponification test	+++	-
Flavonoids	Ferric chloride test	-	-
	Sulphuric acid test	-	-
Glycosides	H_2SO_4 test	-	+++
Phenols	Carotenoids test	-	-
	Potassium dichromate test	-	-
Phytosterols	Libermann-Burchard's test	-	-
	Salkowski's test	+++	++
Protein and Amino Acid	Xanthoproteic test	++	-
Quinones	Sulfuric acid test	+++	++
Saponins	Sodium bicarbonate test	-	+++
Tannins	Lead acetate test	-	-
Terpenoids	Sulfuric acid test	-	-
	Chloroform test	-	+++

were detected in both aqueous and methanol extracts, with the methanol extract showing a stronger presence. Alkaloids are known for their analgesic, anti-inflammatory, and anti-cancer activities, making *Momordica charantia* seeds a valuable source of bioactive compounds for therapeutic purposes. The tests for anthraquinones and emodins were negative, indicating that these compounds, which are often associated with antimicrobial and laxative effects, are absent in the seed extracts. Similarly, no carbohydrates were detected, suggesting that the seeds do not contain significant amounts of simple sugars, which typically provide energy. Although cardiac

glycosides were absent in both extracts, coumarins were present in both the aqueous and methanol extracts. Coumarins are known for their anti-inflammatory, anticoagulant, and antimicrobial properties, indicating that the seeds may possess these beneficial characteristics. The fixed oils and fats in the aqueous extract showed a strong presence, suggesting that the seeds may serve as a source of lipids with potential benefits for skin health and anti-inflammatory processes. The presence of flavonoids was not detected in either extract, which limits the seed's potential antioxidant and cardiovascular protective effects. However, the methanol extract tested positive for glycosides, which are involved in various physiological processes, including anti-inflammatory and antidiabetic activities. Testing for phenols did not yield positive results, indicating the absence of significant phenolic compounds, which are known for their antioxidant properties. On the other hand, both extracts tested positive for phytosterols, with the aqueous extract showing a stronger reaction. Phytosterols are known for their cholesterol-lowering effects and potential anti-inflammatory benefits, making *Momordica charantia* seeds a promising source of these compounds. The protein content in the aqueous extract was confirmed through the Xanthoproteic test, suggesting that proteins are present and contribute to the seed's biological functions. Quinones, which are known for their antimicrobial and anticancer properties, were also detected in both extracts, with a stronger presence in the aqueous extract. Saponins were detected in the methanol extract, indicating their presence as well. Saponins are associated with cholesterol-lowering, antimicrobial, and immune-boosting effects, highlighting the potential therapeutic value of the methanol extract of *Momordica charantia* seeds. Tests for tannins yielded negative results, suggesting that the seeds do not possess significant astringent or antioxidant properties related to tannins. Lastly, terpenoids were present in the methanol extract, indicating the potential of the seeds to exhibit anti-inflammatory, antimicrobial, and anticancer activities. The aqueous extract did not show the presence of terpenoids, suggesting that methanol may be a more effective solvent for extracting these compounds.

GC-MS Analysis

The GC-MS analysis of the methanolic extract of bitter gourd (*Momordica charantia*) seeds identified a diverse range of bioactive compounds, each exhibiting significant biological activities. These compounds, including antimicrobial, anti-inflammatory, antioxidant, and enzyme-inhibitory effects, hold potential for therapeutic applications in treating a variety of diseases. Among the compounds (Table. 2) identified, Methyl valerate ($C_6H_{12}O_2$) with a retention time of 2.371 and a peak area of 1.55% displayed insecticidal properties, offering a potential alternative to synthetic pesticides. It also holds promise in the fragrance and flavor industries. Propanedioic acid, propyl ($C_6H_{10}O_4$), with a retention time of 2.797 and a peak area of 10.35%, showed strong antimicrobial and anti-inflammatory properties. It also acts as an enzyme inhibitor, suggesting its potential use in drug development for treating inflammatory diseases. Oxime, methoxy-phenyl ($C_8H_9NO_2$), with a retention time of 2.931 and a peak area of 1.13%, demonstrated antimicrobial, anti-inflammatory, and antioxidant activities (Figure. 9).

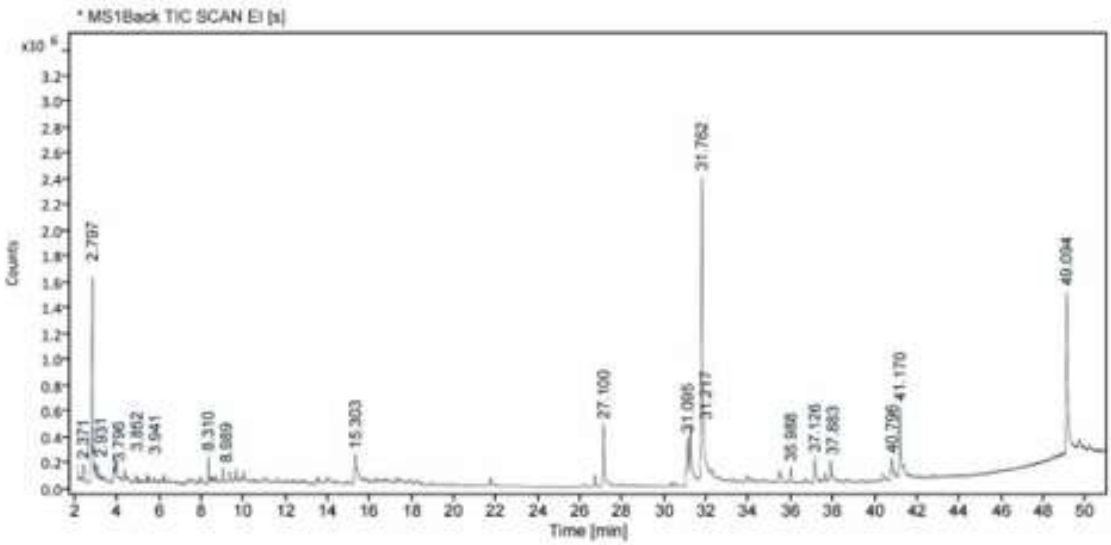

Fig. 9: GC-MS chromatogram of *Momordica charantia* L. seeds in methanolic extract

This compound could be valuable in the development of drugs targeting infections and chronic inflammation. Similarly, 2(3H)-Furanone, dihydro-5-methyl ($C_5H_8O_2$) with a retention time of 3.796 and a peak area of 0.28% and 1,2,6-Hexanetriol ($C_6H_{14}O_3$) with a retention time of 3.852 and a peak area of 0.48%, both exhibited antimicrobial and anti-inflammatory properties, suggesting their utility in treating bacterial infections and inflammation. The compound β-D-Glucopyranose ($C_6H_{12}O_6$), with a retention time of 3.941 and a peak area of 0.37%, was identified for its antioxidant activity and potential as a prebiotic. This compound could be used to promote the growth of beneficial gut bacteria, benefiting overall health. Another significant compound, 2-Heptenal, 2-propyl ($C_{10}H_{18}O$), with a retention time of 8.310 and a peak area of 1.78%, exhibited antimicrobial, anti-inflammatory, and antioxidant properties, making it a valuable candidate for treating infections and oxidative stress. 2,4-Dodecadienal ($C_{10}H_{16}O$), with a retention time of 8.989 and a peak area of 1.00%, showed antimicrobial, antifungal, and insecticidal properties. These characteristics could potentially be harnessed for pest control applications. D-Allose ($C_6H_{12}O_6$), a rare sugar with a retention time of 15.303 and a peak area of 4.24%, demonstrated antioxidant and anti-inflammatory effects. Additionally, it was found to have immunomodulatory effects, suggesting its potential in immunotherapy. The derivative of vitamin C, L-(+)-Ascorbic acid 2,6-dihexadecanoate ($C_{38}H_{68}O_8$), with a retention time of

Table No. 4: Phytochemical Compounds (GC-MS) of *Momordica charantia* seed using Methanol Extract.

Peak	**RT**	**Area %**	**Name of the Compound**	**Molecular Formula**	**Molecular Weight** (g /mol)	**IUPAC Name**
1	2.371	1.55	Methyl valerate	$C_6H_{12}O_2$	116.16	methyl pentanoate
2	2.797	10.35	Propanedioic acid, propyl	$C_6H_{10}O_4$	146.14	2-propylpropanedioic acid
3	2.931	1.13	Oxime, methoxy-phenyl	$C_8H_9NO_2$	151.16	methyl (Z)-N-hydroxybenzenecarboximidate

4	3.796	0.28	2(3H)-Furanone, dihydro-5-methyl	$C_5H_8O_2$	100.12	5-methyloxolan-2-one
5	3.852	0.48	1,2,6-Hexanetriol	$C_6H_{14}O_3$	134.17	hexane-1,2,6-triol
6	3.941	0.37	β-D-Glucopyranose	$C_6H_{12}O_6$	180.16	(2S,3S,4S,5R)-2,3,4,5-Tetrahydroxy-6-(hydroxymethyl)oxane-3-ol
7	8.310	1.78	2-Heptenal, 2-propyl	$C1_0H_{18}O$		(Z)-2-propylhept-2-enal
8	8.989	1.00	2,4-Dodecadienal	$C_{12}H_{20}O$	180.29	(2E,4E)-dodeca-2,4-dienal
9	15.303	4.24	D-Allose	$C_6H_{12}O_6$	180.16	(2R,3R,4R,5S,6R)-6-(hydroxymethyl)oxane-2,3,4,5-tetrol
10	27.100	7.52	l-(+)-Ascorbic acid 2,6-dihexadecanoate	$C_{38}H_{68}O_8$	652.9	[(2S)-2-[2R)-4-hexadecanoyloxy-3-hydroxy-5-oxo-2H-furan-2-yl]-2-hydroxyethyl] hexadecanoate
11	27.100	6.11	9,12-Octadecadienoic acid (Z, Z)	$C_{18}H_{32}O_2$	280.4	(9E,12Z)-octadeca-9,12-dienoic acid
12	31.217	7.19	6-Octadecenoic acid	$C_{18}H_{34}O_2$	287.42	(E)-(1,2,3,4,5-13C5)octadec-6-enoic acid
13	31.762	34.5	Octadecenoic acid	$C_{18}H_{34}O_2$	282.5	octadec-9-enoic acid
14	35.988	1.38	Octadecenoic acid, butyl ester	$C_{18}H_{34}O_2$	282.5	(Z)-octadec-9-enoic acid
15	37.126	3.00	Stearic anhydride	$C_{36}H_{70}O_3$	550.9	octadecanoyl octadecanoate
16	37.883	1.92	9- Octadecanoic acid (Z)-, Oxiranylmethyl ester	$C_{21}H_{38}O_3$	338.52	9-Octadecenoic acid (Z)-, 2-(oxiranylmethyl) ester
17	40.796	0.74	Oleic acid, eicosyl ester	$C_{38}H_{74}O$	563	icosyl (Z)-octadec-9-enoate
18	41.170	3.83	Octadecenoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{21}H_{40}O_4$	361.6	(1,1,2,3,3-pentadeuterio-1,3-dihydroxypropan-2-yl) (Z)-octadec-9-enoate
19	49.094	12.58	Glutinol	$C_{30}H_{50}O$	426.7	(3S,6aS,6aS,6bR,8aR,12aR,14aR,14bS)-4,4,6a,6b,8a,11,11,14a-octamethyl-1,2,3,6,6a,7,8,9,10,12,12a,13,14,14b-

						tetradecahydropicen-3-ol

27.100 and a peak area of 7.52%, exhibited antioxidant properties, anti-inflammatory effects, and potential applications in skincare due to its skin-protective benefits. 6-Octadecenoic acid ($C_{18}H_{34}O_2$), with a retention time of 31.217 and a peak area of 7.19%, and Octadecenoic acid ($C_{18}H_{34}O_2$), with a retention time of 31.762 and a peak area of 34.5%, both displayed strong antimicrobial, anti-inflammatory, and antioxidant properties. These fatty acids have promising therapeutic potential in managing infections and inflammation. Further compounds such as Octadecenoic acid, butyl ester ($C_{18}H_{34}O_2$), with a retention time of 35.988 and a peak area of 1.38%, and Stearic anhydride ($C_{36}H_{70}O_3$), with a retention time of 37.126 and a peak area of 3.00%, also exhibited antimicrobial, anti-inflammatory, and antioxidant effects, making them valuable in pharmaceutical applications. 9-Octadecanoic acid (Z)-, Oxiranylmethyl ester ($C_{21}H_{38}O_3$), with a retention time of 37.883 and a peak area of 1.92%, and Oleic acid, eicosyl ester ($C_{38}H7_4O$), with a retention time of 40.796 and a peak area of 0.74%, demonstrated similar biological activities, suggesting their potential for treating infections and inflammation. Finally, Octadecenoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester ($C_{21}H_{40}O_4$), with a retention time of 41.170 and a peak area of 3.83%, showed antioxidant, anti-inflammatory, and antimicrobial properties, making it suitable for food preservation and the treatment of oxidative stress-related diseases. Glutinol ($C_{30}H_{50}O$), with a retention time of 49.094 and a peak area of 12.58%, exhibited antiproliferative effects against cancer cells, neuroprotective effects, and antioxidant properties, highlighting its potential in treating cancer and neurodegenerative diseases.

The identification of these 19 compounds from bitter gourd seeds through GC-MS emphasizes the diverse bioactive properties of this plant. The compounds' antimicrobial, anti-inflammatory, antioxidant, and enzyme-inhibitory activities suggest that bitter gourd holds significant promise as a natural source for developing therapeutic agents to address various diseases, including microbial infections, inflammation, cancer, and oxidative stress-related disorders. These findings contribute to the growing interest in the medicinal applications of bitter gourd and its compounds in pharmaceutical and biomedical research.

DISCUSSION

This study investigates the bioactive compounds found in the seeds of *Momordica charantia* (bitter gourd) and explores their potential therapeutic applications. The GC-MS analysis identified a diverse range of compounds, including alkaloids, phytosterols, quinones, saponins, terpenoids, and fatty acids, each contributing significant biological activities such as antimicrobial, anti-inflammatory, antioxidant, and enzyme-inhibitory effects. These findings are in line with previous research highlighting the medicinal properties of bitter gourd, which is traditionally used to treat various ailments (Choudhury *et al*., 2013; Verma & Chandra, 2015). Several compounds identified in this study exhibited strong antimicrobial and anti-inflammatory properties, particularly Methyl valerate and Propanedioic acid, propyl, which suggest their potential in treating infections and chronic inflammation. The discovery of Glutinol, a compound with neuroprotective and anticancer properties, further underscores the therapeutic value of bitter gourd seeds, particularly in developing treatments for neurodegenerative diseases and cancer (Wu *et al*., 2010; Lee *et al*., 2012). Additionally, D-Allose, a rare sugar,

demonstrated immunomodulatory effects, further supporting its role in immune-related diseases (Matsuo *et al*., 2002). The antioxidant properties of compounds like β-D-Glucopyranose and L-(+)-Ascorbic acid 2,6-dihexadecanoate provide additional support for bitter gourd seeds in managing oxidative stress, a key factor in the development of chronic diseases such as cardiovascular diseases and cancer (Zhang *et al*., 2013). The anti-inflammatory and antioxidant activities observed in this study suggest that bitter gourd seeds could serve as a natural alternative to synthetic drugs, especially for conditions involving oxidative damage and chronic inflammation.

This study highlights the therapeutic potential of *Momordica charantia* seeds. Further research into the isolation and clinical testing of these bioactive compounds could lead to the development of novel natural therapies for various diseases, including infections, inflammation, cancer, and oxidative stress-related disorders.

CONCLUSION

The seeds of *Momordica charantia* (bitter gourd) are a rich source of bioactive compounds, including alkaloids, phytosterols, quinones, saponins, and terpenoids, each of which contributes to their well-known medicinal properties. These compounds are believed to play a significant role in the plant's therapeutic potential, particularly in the treatment of various health conditions. The presence of these bioactive substances supports the use of bitter gourd seeds in the development of natural therapeutic agents, including those aimed at addressing inflammation, microbial infections, and cholesterol-related disorders.

Furthermore, the GC-MS analysis conducted on *Momordica charantia* seeds identified 19 key bioactive compounds with significant biological activities. These compounds exhibited a variety of beneficial properties, such as antimicrobial, anti-inflammatory, antioxidant, and enzyme-inhibitory effects. The results underscore the plant's potential for treating a broad range of health issues, including infections, chronic inflammation, cancer, and diseases linked to oxidative stress. Notably, compounds like Methyl valerate, Propanedioic acid, propyl, D-Allose, and Glutinol, which were identified in this study, highlight the diverse pharmacological potential of bitter gourd. These findings make it evident that *Momordica charantia* seeds could serve as a valuable source of natural compounds for the development of effective drugs. Their diverse medicinal properties not only support their use in treating multiple diseases but also open exciting opportunities for further exploration in drug development, as well as potential applications in the pharmaceutical and cosmetic industries. Given the therapeutic promise of these bioactive compounds, further research is essential to better understand their specific mechanisms of action and clinical efficacy, paving the way for the future use of bitter gourd as a natural alternative in modern medicine.

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