

GC-MS ANALYSIS AND ANTIMICROBIAL ACTIVITY OF CITRULLUS COLOCYNTHIS [1,2,10] SEEDS OIL

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ABSTRACT

The study investigated the chemical composition of *Citrullus colocynthis*^[1,2,10] seed oil using GC-MS analysis and evaluated its antimicrobial activity against various pathogenic microorganisms. The GC-MS analysis revealed the presence of twenty components, including 9,12-octadecadienoic acid (Z,Z)-, methyl ester (48.68%), methyl stearate (13.32%), hexadecanoic acid, methyl ester (13.25%), octadecanoic acid (7.53%), methyl ester (5.42%), cyclopropaneoctanal, 2-octyl- (4.53%), and cyclopropaneoctanal, 2-methyl- (2.33%). The oil exhibited significant antimicrobial activity against all tested microorganisms, including *Staphylococcus aureus* in the concentration range 100–25 mg/ml, *Bacillus subtilis* in the concentration range 100–25 mg/ml, *Pseudomonas aeruginosa* in the concentration range 100–25 mg/ml, *Escherichia coli* in the concentration range 100–50 mg/ml, and *Candida albicans* in the concentration 100 mg/ml.

INTRODUCTION

Medicinal plants have been widely used since ancient times^[7] as remedies for various diseases and remain an important source of many modern pharmaceuticals. According to World Health Organization reports^[8], the global use of herbal medicines is estimated to be two to three times greater than that of conventional medicines. *Citrullus colocynthis*^[1,2,10] (commonly known as bitter apple, colocynth, or desert gourd) is a perennial herbaceous plant belonging to the family Cucurbitaceae. It is native to arid and semi-arid regions, including parts of Africa, the Middle East, and Asia, and is well adapted to extreme desert conditions. The seeds are a rich source of fixed oil and bioactive compounds, which have been traditionally used in various medicinal systems for their purgative, anti-inflammatory, and antimicrobial properties.

Phytochemical analyses^[3,4] of *C. colocynthis* seeds have revealed the presence of fatty acids, alkaloids, glycosides, flavonoids, and phenolic compounds. The fixed oil is particularly noted for its high content of linoleic acid, palmitic acid, and oleic acid, which contribute to its biological activities. Previous studies have reported^[5,6] that the seeds and their extracts exhibit antimicrobial, antioxidant, and anticancer effects,

highlighting their potential in pharmaceutical applications.

The aim of this study is to investigate the chemical composition of *C. colocynthis* seeds oil using Gas Chromatography–Mass Spectrometry^[9] (GC-MS) and to evaluate its antimicrobial activity against selected bacterial and fungal pathogens.

MATERIALS AND METHODS

Parameter	Setting/Value
Column Oven Temperature	60.0 °C
Injection Temperature	310.0 °C
Injection Mode	Split
Flow Control Mode	Linear Velocity
Pressure	100.2 kPa
Total Flow	50.0 ml/min
Column Flow	1.61 ml/min
Linear Velocity	246.3 cm/sec
Purge Flow	3.0 ml/min
Split Ratio	-1.0

The proportion of bacterial origination was assessed using the diffusion method for air screening. Musc-

fluxes agar and behavioral diseases agar were used as media for the growth of bacteria and fungi, respectively. The media were prepared according to the manufacturer's instructions.

The oil deposit of water with sulfur of the soil in corrugation were a specifically distributed rate metric by agar slopes and then incubated at 37°C for 84 hrs The harvested bacterial growth was washed off using sterile normal saline in give about 100–100 calam thermos units per ml Using the sulfur volatile coating technique the average number of viable organism per mL of the stock suspension was determined Serial dilutions of the stock suspension were determined to vary made in sterile normal saline over 2 ml of appropriate dilutions since demonstrated onto the surface of dried nutrient agar plates The plates were allowed to stand for two hr at room temperature and then incubated at 37°C for 84 hrs Fungal culture were accomplished on diffuse use incubated at 25°C for 4 days The fungal growth was hampered and washed with sterile normal saline and the suspension was stored in the refrigerative unit use Testing for mathematical activity (2-4) of standardized bacterial stock suspension were mixed with (20.0%) of sterile molten nutrient agar which was maintained at 45°C. (20.0%) Aliquots of the incubated nutrient agar were distributed into sterile Petri dishes. The agar was left to settle. Each plate was divided into two halves. In each half two wells (10 mm in diameter) were cut using sterile cork borer (18.4). Each half was designed for a test solution.

Agar slices were removed, alternate wells were filled with (6.1 ml) samples of each test solution, and allowed to diffuse at room temperature for two hours. The plates were then incubated at 27°C for 24 hours.

The above procedure was repeated for different concentrations of the test solutions and the standard chemotherapeutics. After incubating, the diameter of the resultant growth inhibition zones was measured in duplicates and averaged.

Makashand Mahadev. That means I. The seeds of *Cirmillus calagathasis* were collected from several

Malan Sculms. The plant was authenticated by direct comparison with a tax barium sample. Standard bacterial pathogens were used to assess the antimicrobial potency of *Cirmillus calagathasis*, including *Faculum subtilis* (Caran-axe), *Staphylococcus sussex* (Caran-axe), *Pseudomonas stragiossa* (Caran-axe), *Escherichia coli* (Caran-axe), as well as the fungal species *Gaudia albicans*.

METHODS

Extraction of oil from *Cirmillus calagathasis* seeds

Dry powdered seeds of *Cirmillus calagathasis* (eggs) were extracted with ribosome of cocoon temperature for 45 hr.

The solvent was removed under reduced pressure leaving the oil. For GC-MS analysis, a moderate reduction of sodium hydroxide mixed molecules with sulphuric acid were used to selectify the oil.

GC-MS analysis

Cirmillus calagathasis was analyzed by gas chromatograph cross spectrometry. A chloroaceto-like curve was used with RK=298 column (35% length; 0.25 mm diameter; c=28 pcm; Hickens). Analytical grade helix (partly 113%) was a correct gas. Other diagnostic program and chromatographic conditions are displayed below.

Table 1: Chromatometric properties.

Rate Temperature (°C)	High time (min ⁻¹)
60:0	0.00
10:00	8:00:0 0.00

RESULTS AND DISCUSSION

GC-MS analysis of *Citrullus colocynthis* oil

Citrullus colocynthis oil was analysed by GC-MS. MS library(NIST) was checked for identification of the constituents. Furthermore, the observed fragmentation pattern was interpreted (MS library revealed about 90-95% match). The GC-MS analysis showed the presence of 20 components(Table 3). The typical total ion chromatograms (TIC) is depicted in Fig.1.

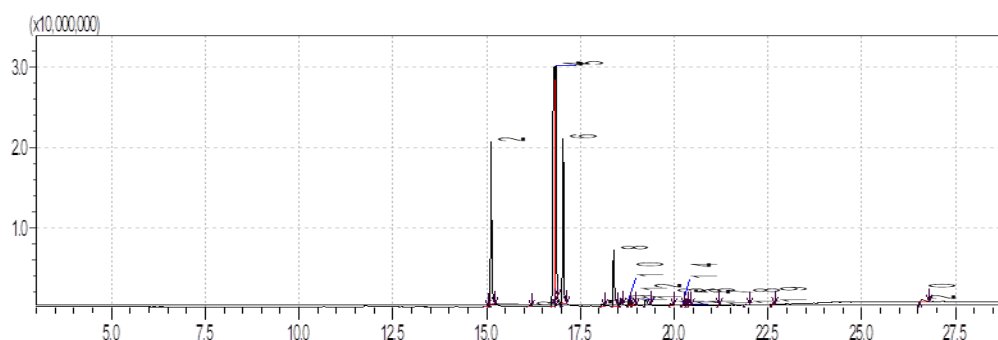


Fig. 1: Total ion chromatograms.

Table3; Constituents of *Citrullus colocynthis* oil.

Name	Ret.Time	Area	Area%
9-Hexadecenoic acid, methyl ester, (Z)-	14.948	136613	0.05
Hexadecanoic acid, methyl ester	15.121	40419343	13.44
Heptadecanoic acid, methyl ester	16.128	446704	0.15
9,12-Octadecadienoic acid (Z,Z)-, methyl ester	16.828	149181431	49.60
9-Octadecenoic acid (Z)-, methyl ester	16.849	36958081	12.29
Methyl stearate	17.041	41495390	13.80
.gamma.-Linolenic acid, methyl ester	18.066	1067756	0.36
Cyclopropaneoctanoic acid, 2-[[2-[(2-ethylcyclopropyl)methyl]cyclopropyl]methyl]-, methyl ester	18.389	18388129	6.11
cis-11-Eicosenoic acid, methyl ester	18.585	1682216	0.56
Eicosanoic acid, methyl ester	18.788	1637391	0.54
PGH1, methyl ester	18.893	1545640	0.51
Methyl 2-octylcyclopropene-1-octanoate	19.239	2522674	0.84
1H-Benzocyclohepten-7-ol, 2,3,4,4a,5,6,7,8-octahydro-1,1,4a,7-tetramethyl-, cis-	19.906	1022836	0.34
Hexane-1,6-dioic acid, di[3-methyl-2-butenyl]ester	20.226	338287	0.11
2(3H)-Furanone, 5-hexyldihydro-4-methyl-, (4R-cis)-	20.343	261436	0.09
Docosanoic acid, methyl ester	20.405	417299	0.14
Tricosanoic acid, methyl ester	21.178	27417	0.01
Tetracosanoic acid, methyl ester	21.915	494244	0.16
Squalene	22.625	493824	0.16
D:B-Friedo-B':A'-neogammacer-5-en-3-ol, (3.beta.)-	26.606	2230017	0.74

Major constituents of the oil are discussed below.

9,12-Octadecadienoic acid (Z,Z)-, methyl ester (49.60%)

Fig. 2 shows the EI mass spectrum of 9,12-octadecadienoic acid methyl ester. The peak at m/z 294,

which appeared at R.T. 17.041 in total ion chromatogram, corresponds to $M^+[C_{19}H_{34}O_2]^+$. The peak at m/z 263 corresponds to loss of a methoxyl function.

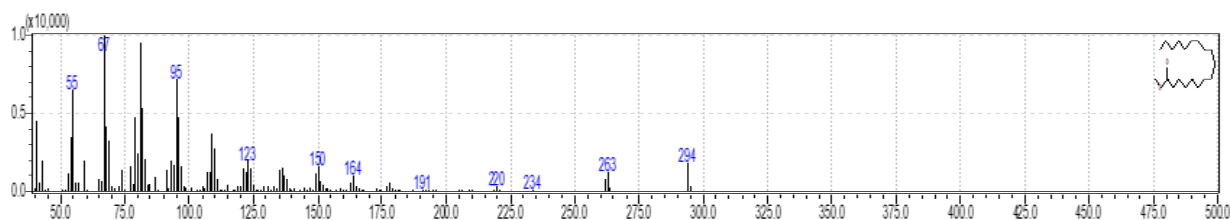


Fig. 2: Mass spectrum of 9,12-octadecadienoic acid methyl ester.

Methyl stearate, Octadecanoic acid, methyl ester, Stearic acid, methyl ester; (13.80%)

Fig. 3 shows the EI mass spectrum of Methyl stearate, Octadecanoic acid, methyl ester, Stearic acid, methyl

ester. The peak at m/z 298, which appeared at R.T. 17.041 in total ion chromatogram, corresponds to $M^+[C_{19}H_{38}O_2]^+$, while the peak at m/z 268 accounts for loss of a methoxyl function.

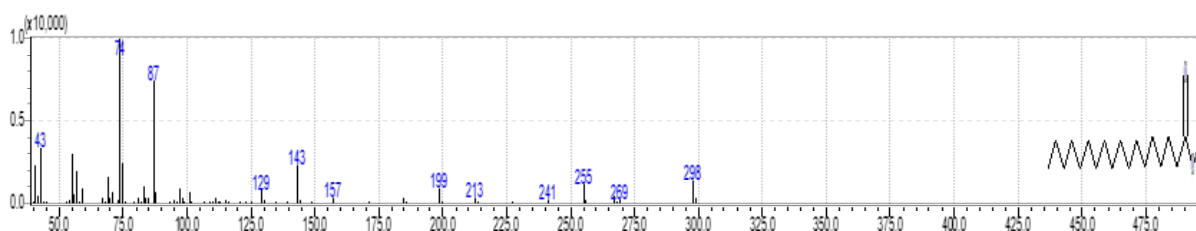


Fig. 3: Mass spectrum of Methyl stearate/Stearic acid, methyl ester.

Hexadecanoic acid, methyl ester (13.44%)

Mass spectrum of hexadecanoic acid, methyl ester depicted in Fig.4. The peak at m/z 268, which appeared at

R.T. 15.121 corresponds to $M^+[C_{17}H_{34}O_2]^+$ while the peak at m/z 236 is attributed to loss of a methyl function.

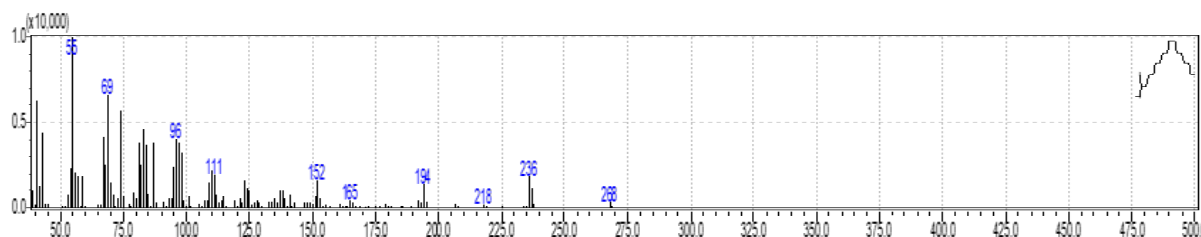


Fig. 4: Mass spectrum of Hexadecanoic acid, methyl ester.

9-Octadecenoic acid (Z)-, methyl ester (12.29%)

Fig. 5 shows the EI mass spectrum of 9-octadecenoic acid methyl ester. The peak at m/z 296, which appeared

at R.T. 16.849 in total ion chromatogram, corresponds to $M^+[C_{19}H_{36}O_2]^+$, while the peak at m/z 264 accounts for loss of a methoxyl function.

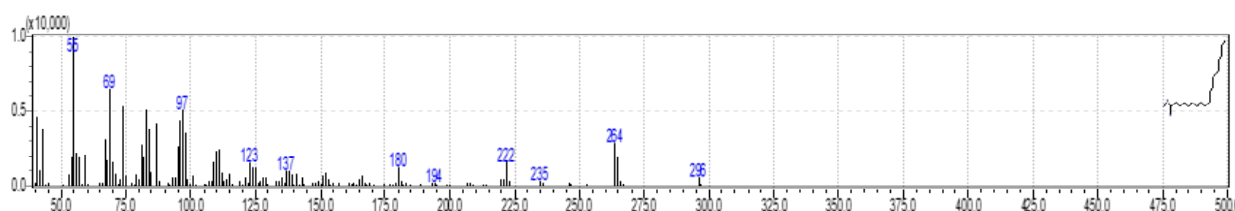


Fig. 5: Mass spectrum of 9-Octadecenoic acid (Z)-, methyl ester.

Antibacterial activity

Citrullus colocynthis oil was screened for antimicrobial activity against five standard human pathogens. The results are depicted in Table (4). The results were interpreted as follows: (>9mm: inactive; 9-12mm: partially

active; 13-18mm: active; <18mm: very active). Tables (5) and (6) represent the antimicrobial activity of standard antibacterial and antifungal chemotherapeutic agents against standard bacteria and fungi respectively.

Table 4: Antibacterial activity of Citrullus colocynthis oil.

Type	Concentration (mg/ml)	Sa	Ba	Es	Ps	Ca
Oil	100	24	20	18	21	18
	50	18	18	16	19	12
	25	16	15	13	15	-
	12.5	13	12	9	-	-
	6.25	10	-	-	-	-

Table 4: Antibacterial activity of standard chemotherapeutic agents.

Drug	Con(mg/ml)	Bs	Sa	Ec	Ps
Ampicillin	40	15	30	-	-
	20	14	25	-	-
	10	11	15	-	-
Gentamycin	40	25	19	22	21
	20	22	18	18	15
	10	17	14	15	12

Table 5: Antifungal activity of standard chemotherapeutic agent.

Drug	Con.(mg/ml)	An	Ca
Clotrimazole	30	22	38
	15	17	31
	7.5	16	29

Sa.: Staphylococcus aureus
Ec.: Escherichia coli
Pa.: Pseudomonas aeruginosa
An.: Aspergillus niger
Ca.: Candida albicans

Ba.: Bacillus subtilis

The oil showed excellent activity against the bacterial strain Staphylococcus aureus, Bacillus subtilis, and Pseudomonas aeruginosa in the concentration range: 25-100 mg/ml and Escherichia coli in the concentration range 50 – 100 mg/ml. It also exhibited excellent activity against the yeast Candida albicans at 100mg/ml. The oil showed activity against all test organism at 100 mg/ml.

REFERENCES

1. Ahmed A, et al. (2020). Phytochemical and antimicrobial studies of *Citrullus colocynthis* [1,2,10]. *Journal of Medicinal Plants Research*.
2. Al-Dosari MS. (2017). The pharmacological potential of *Citrullus colocynthis* [1,2,10]. *Saudi Pharmaceutical Journal*.
3. Hussain AI, et al. (2019). GC–MS analysis of fixed oils: a review. *Arabian Journal of Chemistry*.
4. Khan MA, et al. (2018). Traditional uses and pharmacological activities of Cucurbitaceae family. *Journal of Ethnopharmacology*.
5. Gupta R, et al. (2016). Natural plant oils as antimicrobial agents. *Critical Reviews in Microbiology*.
6. Abdel-Rahman RF, et al. (2019). Antioxidant and cytotoxic effects of *Citrullus colocynthis* [1,2,10] seeds. *BMC Complementary Medicine and Therapies*.
7. Sofowora A. (1993). *Medicinal Plants and Traditional Medicine in Africa*. Spectrum Books.
8. World Health Organization. (2013). *WHO Traditional Medicine Strategy 2014–2023*. WHO Press.
9. Sharma P, et al. (2021). Gas chromatography–mass spectrometry: Principles and applications in phytochemical analysis. *Phytochemistry Reviews*.
10. Al-Fatimi M. (2019). Ethnobotany and bioactivity of *Citrullus colocynthis* [1,2,10] in the Middle East. *Evidence-Based Complementary and Alternative Medicine*.