



IDENTIFICATION OF BIOACTIVE PHYTOCHEMICALS IN *MOMORDICA CHARANTIA* FRUIT METHANOLIC EXTRACT BY GC-MS: INSIGHTS INTO POTENTIAL BIOLOGICAL ACTIVITIES

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ABSTRACT

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This study discusses the bioactive composition of the methanolic extraction and the biological activities of *Momordica charantia*. Dried fruit slices of 100% pure organic *Momordica charantia* fruit were used to prepare a methanolic extract using by Soxhlet extractor device. The GC-MS Profiling was performed. The separation product was analyzed based on the resulting mass spectra and compared with a database of reference spectra (NIST20.L) library to identify the chemical compounds present in the extract. Different chemical constituents were contributed to the final phytochemical profile, which enhances the phytochemical profiles identified in previous studies of *Momordica charantia* fruit, the major constituents in this study included: 6-pentylpiperidin-2-one; 1,4-Benzenedicarboxylic acid, bis(2-ethylhexyl) ester; Myo-Inositol, 4-C-methyl-; 1-Methyl-5-fluorouracil; 1,3,5,7-Tetroxane; Butanenitrile, 4-(dimethylamino)-; 1,4-Butanediamine, 2,3-dimethoxy-N, N, N', N'-tetramethyl-, [S-(R*,R*)]-; Propane, 2-fluoro-2-methyl-; n-Hexadecanoic acid; Phenethyl alcohol, 2,5-dihydroxy-.alpha.-methyl-, (-)-; and Maltol. The potential components and their derivatives were confirmed to have important biological effects for health, such as antibacterial, anticancer, antioxidants, anti-inflammatory, antimicrobial, hepatoprotective, antiviral, antimalarial, antipyretic, and antitumor applications. These biological effects of the methanolic extract of *Momordica charantia* fruit can directly affect human health. The results of this study are essential, as they can inform subsequent research into the extraction of bioactive components from *Momordica charantia* and their uses.

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INTRODUCTION

Momordica charantia [Family: *Cucurbitaceae*; Genus: *Momordica*] Common Names: Bitter-melon, Bitter-gourd, Bitter-squash, karela, and Karavella derived from Sanskrit. It is a medicinal plant utilized as a vegetable and traditional medicine exhibiting hypoglycemic properties in the fruits (Singh *et al.*, 2011; Joseph and Jini, 2013). The fruit of *Momordica charantia* is bright orange, measuring 5-15 cm in length, pendulous, fusiform, typically pointy or beaked, ribbed, and adorned with numerous triangular tubercles that resemble a crocodile's back, featuring three valves at the tip during maturity (Çetintaş *et al.*, 2021).

Bitter melon, a fruit of the tropical region, belongs to the class Magnoliopsida, order *Cucurbitales*, family *Cucurbitaceae*, and genus *Momordica*, with the Latin

designation *Momordica charantia*. It is also referred to as strange apple, bitter apple, miracle apple, bitter melon, papara (Çetintaş *et al.*, 2021).

It is a tropical and subtropical vine, extensively cultivated in Asia, Africa, and the Caribbean for its edible fruit, among the most bitter fruits. It thrives in tropical regions, serving as sustenance and therapeutic agents. It is a perennial vine with elongated stalked leaves that yield solitary yellow male and female flowers located in the leaf axils. The fruit is a warty gourd, typically oblong and resembling a little cucumber. The immature fruit is vibrant green, transitioning to orange-yellow upon ripening. Upon ripening, the fruit divides into three uneven valves that curl backward, dispersing countless brown or white seeds encased in crimson arils. The generic name "Momordica" is derived from Latin, signifying "to bite." Ripe fruits exhibit greater bitterness than unripe fruits, attributed to cucurbitacin-like alkaloid momordicine and triterpene glycosides. To reduce the bitterness of bitter gourd, apply salt uniformly to the pieces and allow them to rest for 30 minutes. This will mitigate the bitter flavor of the gourd, rendering it more pleasant, or alternatively, immerse it in tamarind juice for several minutes (Sharma, 2019).



Indian Bitter-melon, Chinese Bitter-melon, Ripe fruit of Bitter Gourd

Figure (1): *Momordica charantia* Fruit (Adapted from Sharma, 2019, A chemical and medicinal potency of *Momordica charantia*. The Pharma Innovation Journal, 2019; 8(6):531-536)

As fruits mature, they will exhibit a delicate texture, a pale green color, and a rough or uneven exterior. Permit maturation until they attain an orange color and thereafter rupture to disclose numerous seeds (Sharma, 2019).

The primary components of *Momordica charantia* are triterpene, steroid, alkaloid, protein, lipid, and phenolic compounds (Gupta *et al.*, 2011). Despite their bitterness, the fruits of bitter gourd are nutritious and valued as a vegetable when immature. They are utilized medicinally for their acrid, anti-diabetic, anti-inflammatory, digestive, purgative, stimulant, stomachic, and appetite-enhancing properties. They can be sliced and dried for preservation and use during the off-season (Sharma, 2019).



8-13 mm long, compressed, corrugate on the margin, sculptured on both faces

Figure (2): Ripe split fruit of *Momordica charantia* (Adapted from Sharma, 2019, A chemical and medicinal potency of *Momordica charantia*. The Pharma Innovation Journal, 2019; 8(6):531-536)



Overripe fruit

Figure (3): Over ripe split fruit of *Momordica charantia* (Adapted from [Pomegranate - Wikipedia, the free encyclopedia](#), 2023)

The fruit of the plant includes zinc, calcium, phosphorus, iron, carotene, thiamine, nicotinic acid, riboflavin, ascorbic acid, copper, and potassium (Patel et al., 2010) and is rich in vitamins A, B, and C (Hamissou *et al.*, 2013).

This plant has been traditionally utilized as a medical food and is claimed to possess therapeutic and antidiabetic properties attributed to its bioactive components (Joseph and Jini, 2013; Kwatra *et al.*, 2016; Tan *et al.*, 2016). It is a fruit that contains nutrients such as potassium, magnesium, and zinc, which serve diverse roles in the body. The fruit of *Momordica charantia*, rich in fiber, is essential for the health of the digestive system. Beta carotene, a carotenoid in bitter melon, is utilized by the body as vitamin A. Vitamin A is crucial for safeguarding vision. The alkaloids in the fruit are the constituents responsible for the bitter flavor of *Momordica charantia*, located in various areas of the plant (Çetintaş *et al.*, 2021). The fruit of *Momordica charantia* is utilized for asthma, burning sensations, constipation, colic, diabetes, cough, fever (malaria), gout, helminthiasis, leprosy, inflammation, skin disorders, ulcers, and wounds. It has been reported to possess hypoglycaemic (antidiabetic) effects in both animal and human investigations. The juice of *Momordica charantia*

leaves is utilized to treat hemorrhoids completely. *Momordica charantia* is utilized as a blood purifier because of its bitter tonic characteristics. It can remedy boils and other hematological issues that manifest on the skin. The juice of *Momordica charantia* is advantageous for treating and preventing liver disease (Ahmad *et al.*, 2016). The ethanolic extract of *Momordica charantia* fruit demonstrated significant efficacy against bacterial infections associated with poultry (Esther *et al.*, 2019). The bitter gourd fruits possess antidiabetic, anti-obesogenic, antimicrobial, antioxidant, anti-inflammatory (de Oliveira *et al.*, 2018; Ravichandiran and Parani, 2021, Pehlivan, 2021), anticancer, anti-helminthic (Ravichandiran and Parani, 2021; Pehlivan, 2021), anti-HIV properties (Ravichandiran and Parani, 2021) and anti-mutagenic (Pehlivan, 2021). Furthermore, the biological impacts of *Momordica charantia* extracts involve neuroprotective, antimalarial, and allelopathic properties (de Oliveira *et al.*, 2018). Recent investigations on the immunoenhancing and immunostimulating activities of *Momordica charantia* have garnered considerable attention and achieved substantial advancements (Wang *et al.*, 2022). In the present work, some *Momordica charantia* phytoconstituents will be determined using GC-MS and focusing on their biological activities.

MATERIALS AND METHODS

Preparation of *Momordica charantia* Fruit Powder

Dried fruit slices of 100% pure organic *Momordica charantia* (bitter melon) from India (Pure Indian Foods), Figure (4) were used in the preparation of the methanolic extract. The seeds were separated from the dried fruit slices, then ground by an electrical grinder.



Figure (4): Dried fruit slices of 100% pure organic *Momordica charantia* Fruit

Preparation of Methanolic Extract of *Momordica charantia*

50g of the powder of dried fruit were extracted by methanol (500ml) 70% and double distilled water 30%; to ensure maximum purity and minimize contamination (Double distilled water is preferred because it ensures cleaner, more consistent extraction, prevents unwanted chemical interactions, and maintains the integrity of the methanol-plant system), in soxhlet extractor device at 75-80°C for 13 hours. The final concentrated extracted liquid was kept in an incubator (37°C) until the methanol was evaporated completely (fig. 5). GC-Mass was carried out at Quality Control

Laboratories/Nahran Bin Omar Field/Basra Oil Company to check the extract's constituents.

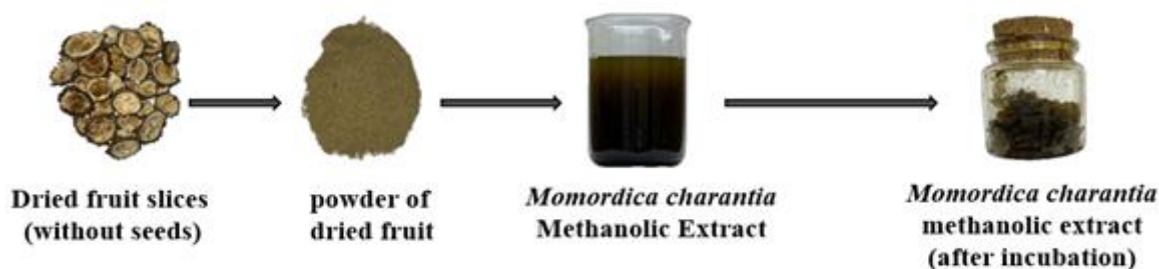


Figure (5): Preparation of Methanolic Extract of *Momordica charantia* Fruit

Phytochemical Profiling of Methanolic Extract of *Momordica charantia* Fruit

The GC-MS Profiling was conducted at Quality Control Laboratories/Nahran Bin Omar Field/Basra Oil Company. The screening of the methanolic extract of *Momordica charantia* fruit was performed by Gas Chromatography coupled to mass spectrometry (GC-MS) to identify the active chemical compounds. The sample was injected into the separation column under specific operating conditions, including temperature and a gradual heating program. The separation product was analyzed based on the resulting mass spectra and compared with a database of reference spectra (NIST20.L) library to identify the chemical compounds present in the extract. This technique helps in identifying active components that may have useful biological properties.

RESULTS AND DISCUSSION

The methanolic extract of *Momordica charantia* fruit was analyzed using GC-MS. The Retention Time (RT), chemical name, molecular formula, CAS ID, and relative abundance (Area%) of each compound were recorded and matched with library data (NIST20.L) for identification. Several compounds were identified in the extract, as shown in Figure (6). The phytoconstituents identified are shown in the Table (1).

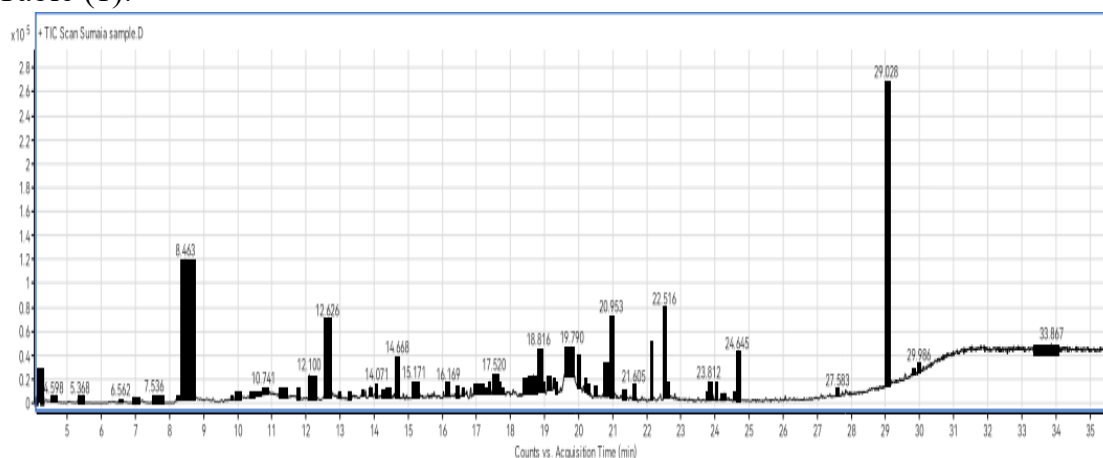


Figure (6): Overlapped GC-MS chromatograms of the methanolic extract of *Momordica charantia* fruit

Table (1): Phytochemical components composition of *Momordica charantia* fruit methanolic extract

No.	RT	Name	Formula	Precursor	Score (Lib)	Area	CAS ID	Area%	Library
1	4.18	Propane, 2-fluoro-2-methyl-	C4H9F	61.02	51.78	123757	353-61-7	3.683486325	NIST20.L
2	4.597	2-Propenoic acid, ethenyl ester	C5H6O2	54.98	55.71	21430	2177-18-6	0.6378395722	NIST20.L
3	6.947	Proline, 2-methyl-5-oxo-, methyl ester	C7H11NO3	98.01	66.56	25560	56145-24-5	0.7607643241	NIST20.L
4	7.534	4(1H)-Pyrimidinone	C4H4N2O	95.97	58.81	29164	4562-27-0	0.8680332843	NIST20.L
5	8.463	6-pentylpiperidin-2-one	C10H19NO	98.01	76.5	848017	1000441-61-5	25.24026134	NIST20.L
6	10	2H-Pyran-2,6(3H)-dione	C5H4O3	56.98	59.73	39728	5926-95-4	1.182458727	NIST20.L
7	10.374	1,3,5,7-Tetroxane	C4H8O4	61.02	42.71	151023	293-30-1	4.495027798	NIST20.L
8	10.741	Butanenitrile, 4-(dimethylamino)-	C6H12N2	58.02	63.41	139691	13989-82-7	4.157743709	NIST20.L
9	10.859	1,4-Butanediamine, 2,3-dimethoxy-N,N,N',N'-tetramethyl-, [S-(R*,R*)]-	C10H24N2O2	58.02	62.05	134918	26549-21-3	4.015680793	NIST20.L
10	12.104	Maltol	C6H6O3	126	68.59	69617	118-71-8	2.072070812	NIST20.L
11	12.622	1-Methyl-5-fluorouracil	C5H5FN2O2	144	77.3	216258	1000427-92-0	6.436673365	NIST20.L
12	13.485	N-Ethyl-2-isopropoxycarbonylazetidine	C9H17NO2	83.98	56.72	37235	54773-06-7	1.108257418	NIST20.L
13	13.896	3-Pyridinecarbonitrile, 1,4-dihydro-1-methyl-	C7H8N2	118.97	68.88	24672	19424-15-8	0.7343340142	NIST20.L
14	14.072	p-Acrylotoluidide	C10H11NO	160.98	50.06	12327	7766-36-1	0.3668991324	NIST20.L
15	14.67	1H-Indene, 2,4,5,6,7,7a-hexahydro-7a-methyl-3-(2-methylpropyl)-	C14H24	191.98	71.55	32928	66708-26-7	0.9800644626	NIST20.L
16	15.17	4-Acetoxy-3-methoxystyrene	C11H12O3	150.01	80.04	23009	46316-15-8	0.684836711	NIST20.L
17	17.674	N-[(1-Ethyl-2-pyrrolidinyl)methyl](2-fluorophenyl)methanamine	C14H21FN2	98.01	42.07	42204	726162-88-5	1.256154051	NIST20.L
18	18.811	4-(4-Ethylphenyl)butanoic acid	C12H16O2	134.05	58.12	49634	5467-53-8	1.477299549	NIST20.L
19	19.098	1,3-Benzenediol, o-(4-ethylbenzoyl)-o'-(3-methylbenzoyl)-	C23H20O4	159.07	48.9	17498	1000330-83-6	0.5208080651	NIST20.L
20	19.262	1,2-Benzenediol, O-propoxycarbonyl-O'-(2-(trifluoromethyl)benzoyl)-	C18H15F3O5	173.01	46.96	11297	1000329-75-6	0.336242354	NIST20.L
21	19.688	Butanoic acid, 2-oxo-, methyl ester	C5H8O3	59.02	58.32	29203	3952-66-	0.869194075	NIST20.L
22	19.79	Myo-Inositol, 4-C-methyl-	C7H14O6	87.01	67.03	404713	472-95-7	12.04582206	NIST20.L
23	20.008	2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5-trimethyl-	C13H22O2	135.02	60.71	43559	36151-02-7	1.296484084	NIST20.L
24	20.776	2-Cyano-N-(4-fluorophenyl)acetamide	C9H7FN2O	111.03	49.89	30062	1735-88-2	0.8947612328	NIST20.L
25	20.952	Phenethyl alcohol, 2,5-dihydroxy-.alpha.-methyl-, (-)-	C9H12O3	124.02	61.1	93544	14293-26-6	2.784230749	NIST20.L
26	21.358	Phosphorus P4	P4	124.02	46.47	13640	1000335-29-2	0.4059790837	NIST20.L
27	21.609	Phenol, 4-propoxy-	C9H12O2	109.95	66.4	11309	18979-50-5	0.3365995204	NIST20.L
28	22.139	Pentadecanoic acid, 14-methyl-, methyl ester	C17H34O2	74.01	70.66	61051	5129-60-2	1.817113566	NIST20.L
29	22.515	n-Hexadecanoic acid	C16H32O2	73.01	71.85	119798	57-10-3	3.565651193	NIST20.L
30	24.031	Methyl stearate	C19H38O2	74.01	46.55	10207	112-61-8	0.303799744	NIST20.L
31	24.651	Butyl citrate	C18H32O7	185.03	75.32	50182	77-94-1	1.493610145	NIST20.L
32	29.028	1,4-Benzenedicarboxylic acid, bis(2-ethylhexyl) ester	C24H38O4	261.13	95.41	409153	6422-86-2	12.17797361	NIST20.L
33	31.98	D-.alpha.-Tocopherol succinate	C33H54O5	282.02	33.07	33391	3/3/4345	0.9938451309	NIST20.L

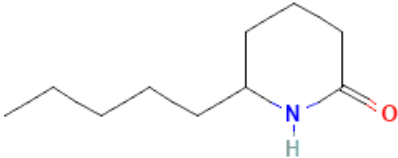
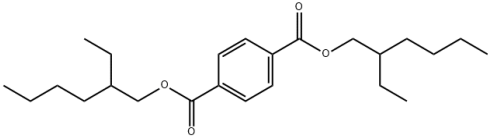
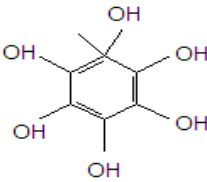
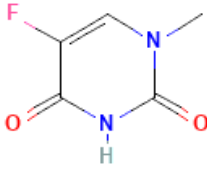
The potential phytoconstituents identified according to peak area% and retention time are shown in Table (2).

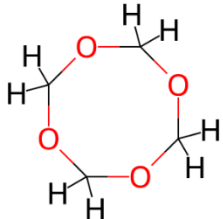
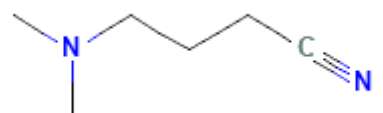
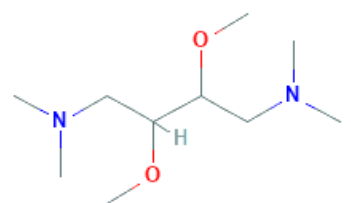

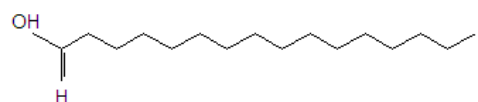
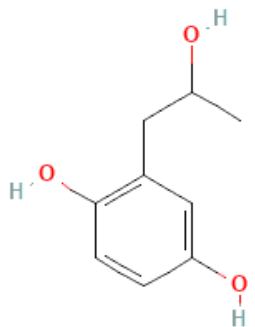
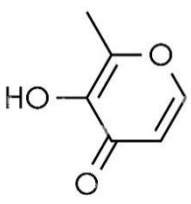
Table (2): Potential phytochemical components composition of *Momordica charantia* fruit methanolic extract

No.	Peak Area%	RT	Phytochemical components
1	25.24026134	8.463	6-pentylpiperidin-2-one
2	12.17797361	29.028	1,4-Benzenedicarboxylic acid, bis(2-ethylhexyl) ester
3	12.04582206	19.79	Myo-Inositol, 4-C-methyl-
4	6.436673365	12.622	1-Methyl-5-fluorouracil
5	4.495027798	10.374	1,3,5,7-Tetroxane
6	4.157743709	10.741	Butanenitrile, 4-(dimethylamino)-
7	4.015680793	10.859	1,4-Butanediamine, 2,3-dimethoxy-N,N,N',N'-tetramethyl-, [S-(R*,R*)]-
8	3.683486325	4.18	Propane, 2-fluoro-2-methyl-
9	3.565651193	22.515	n-Hexadecanoic acid
10	2.784230749	20.952	Phenethyl alcohol, 2,5-dihydroxy-.alpha.-methyl-, (-)-
11	2.072070812	12.104	Maltol

The chemical structures of some of the potential phytoconstituents, components composition of *Momordica charantia* fruit methanolic extract according to their formula are shown in Table (3).

Table (3): Structures of potential phytochemical components composition of *Momordica charantia* fruit methanolic extract

Phytochemical components	Formula	Structure
6-pentylpiperidin-2-one	C ₁₀ H ₁₉ NO	
1,4-Benzenedicarboxylic acid, bis(2-ethylhexyl) ester	C ₂₄ H ₃₈ O ₄	
Myo-Inositol, 4-C-methyl-	C ₇ H ₁₄ O ₆	
1-Methyl-5-fluorouracil	C ₅ H ₅ FN ₂ O ₂	

Phytochemical components	Formula	Structure
1,3,5,7-Tetroxane	C ₄ H ₈ O ₄	
Butanenitrile, 4-(dimethylamino)-	C ₆ H ₁₂ N ₂	
1,4-Butanediamine, 2,3-dimethoxy-N,N,N',N'-tetramethyl-, [S-(R*,R*)]-	C ₁₀ H ₂₄ N ₂ O ₂	
Propane, 2-fluoro-2-methyl-	C ₄ H ₉ F	
n-Hexadecanoic acid	C ₁₆ H ₃₂ O ₂	
Phenethyl alcohol, 2,5-dihydroxy-.alpha.-methyl-, (-)-	C ₉ H ₁₂ O ₃	
Maltol	C ₆ H ₆ O ₃	

6-Pentylpiperidin-2-one is a chemical compound belonging to the class of lactams (cyclic amides). Its structure consists of a piperidin-2-one ring (a six-membered ring containing one nitrogen atom and a carbonyl group at position 2) with a pentyl group attached at the 6-position of the ring. 6-pentylpiperidin-2-one is one of the active compounds of Barhi date seed ethanolic extracts (Shareef, 2025). Itself does not appear to have well-documented direct biological activities in the mainstream literature or databases. However, compounds in the piperidin-2-one family, including substituted derivatives like 6-pentylpiperidin-2-one, are often studied as precursors or scaffolds in medicinal chemistry. They typically synthesize pharmacologically active molecules targeting neurodegenerative diseases, inflammatory conditions, and sometimes bone anabolic agents. So, while 6-pentylpiperidin-2-one might not have specific reported biological activities on its own, its relevance lies mostly in its role as a building block for compounds with biological effects. This is consistent with the general biological activity of piperidin-2-one derivatives, which serve as valuable intermediates in drug development and chemical biology research. No direct evidence specifically shows that 6-pentylpiperidin-2-one has anticancer effects *in vitro* or antioxidant effects *in vivo* documented in the scientific literature or chemical databases.

More broadly, some piperidin-2-one derivatives and related piperidine compounds have been studied for anticancer properties *in vitro* (Romero *et al.*, 2020). However, these are usually modified analogs rather than the simple 6-pentyl substituted version. Similarly, antioxidant effects have been observed with certain piperidine-containing compounds (Manjusha *et al.*, 2018). Therefore, although related molecules show promise, further research is necessary to confirm whether 6-pentylpiperidin-2-one alone exhibits anticancer activity *in vitro* or antioxidant effects *in vivo*.

1,4-Benzenedicarboxylic acid, bis(2-ethylhexyl) ester, (di-(2-ethylhexyl) terephthalate), this chemical was initially isolated by researchers in 1989 and subsequently from various medicinal plants. *Ehretia laevis* (Arbale *et al.*, 2011), *Piper longum* L. Piperaceae (Das *et al.*, 2012), *Catharanthus roseus* (Dhankhar *et al.*, 2014), essential oil derived from *Cirsium japonicum* DC., *Dryopteris ryo-itoana* Kurata, *Aspidium* of the Dryopteridaceae family, and the aerial components of *Dryopteris sublaeta*, together with the leaves of *Panax pseudoginseng* subsp. *himalaicus* and its variants, as well as sunflower varieties var. *angustifolius* and var. *bipinnatifidus*. Additionally, this chemical was extracted from the seahorse *Hippocampus kuda* (Singh *et al.*, 2011). Its biological activities have been studied with mixed findings anticancer activity and insecticidal activity. Some studies have shown positive anticancer effects of this compound on various human cancer cell lines including PC3

(prostate), MCF (breast), HCT-116 (colon), A549 (lung), and MIAPACA (pancreatic) cells, indicating potential cytotoxic effects in vitro (Save *et al.*, 2015).

Myo-Inositol, 4-C-methyl-, the compound was found to be present in major quantity and has been reported as a promising treatment for preventing ovarian hyperstimulation syndrome in experimental rats (Tangavelou *et al.*, 2018). It possesses a wide spectrum of biological activities such as anticancer, anti-inflammatory, antioxidant, antimicrobial, and hepatoprotective (Lin *et al.*, 1996; Dave and Ledwani, 2012; Hsu and Chung, 2012; Dong *et al.*, 2016; Sharma *et al.*, 2017). While specific studies on the 4-C-methyl derivative are limited, myo-inositol is well-documented to exhibit antioxidant and anticoagulant effects (ex vivo) (Rolnik *et al.*, 2024), Broad-spectrum anticancer activity, affecting cell cycle and signaling pathways (Bizzarri *et al.*, 2016).

1-Methyl-5-fluorouracil (1-Me-5-FU) is a methylated derivative of the chemotherapeutic agent 5-fluorouracil (5-FU). Shareef claimed that 1-Methyl-5-fluorouracil is also present as one of the active compounds of Barhi date seed ethanolic extracts (Shareef, 2025). 1-Methyl-5-fluorouracil functions similarly to 5-FU—it's a pyrimidine analog that undergoes intracellular conversion to FdUMP, which forms a covalent complex with thymidylate synthase, blocking the conversion of dUMP to dTMP. This depletion of dTMP leads to inhibited DNA synthesis and induces thymineless death in rapidly dividing cells (Mafi *et al.*, 2023; Voiculescu *et al.*, 2019). Its metabolites (e.g., FUTP, FdUTP) can substitute for uridine or thymidine during RNA/DNA synthesis, causing RNA dysfunction and DNA damage, further promoting cell cycle arrest and apoptosis (Mafi *et al.*, 2023). 5-FU (and likely the methylated form) is effective against cancers of the colon, rectum, stomach, breast, pancreas, and head & neck due to selective toxicity toward proliferating cells (Voiculescu *et al.*, 2019).

1,3,5,7-Tetroxane is one of the active compounds found in Iraqi buffalo buttermilk (Al Musa and Al Garory, 2023; Al Garory and Al Musa, 2024). Multiple studies have described the antimalarial activity of tetraoxane derivatives, including 1,3,5,7-tetroxane, which highlights their peroxide bond crucial for activity against malaria parasites (Vennerstrom *et al.*, 1999; Narula *et al.*, 2019). Antimicrobial and antifungal properties have been reported in research articles and chemical product summaries (Noman and Dehghan, 2022). 1,3,5,7-tetroxane was identified as a major bioactive compound in the *Beta vulgaris* (L.) ethanolic leaf extract, which may contribute to the therapeutic potential as antimalarial, antipyretic, or anti-inflammatory agents (Sargunam and Thilakavathy, 2021). Study by Pavithra, 1,3,5,7-tetroxane was identified as one of the bioactive compounds present in *Artemisia pallens pollen*. The compound was among several phytochemicals detected and was associated with antioxidant properties observed in the extract. This suggests that

1,3,5,7-tetroxane contributes to the antioxidant activity of *Artemisia pallens pollen* (Pavithra *et al.*, 2018).

Butanenitrile, 4-(dimethylamino)- Butanenitrile, 4-(dimethylamino)- regarding anticancer activity, while there is no direct study exclusively on 4-(dimethylamino) butanenitrile itself, related compounds with dimethylamino groups have shown anticancer effects *in vitro* by inhibiting cancer cell colony formation, migration, and tumor spheroid growth, particularly in breast and pancreatic cancer models (Kairyte *et al.*, 2024). As for antioxidant activity, specific data on 4-(dimethylamino) butanenitrile is limited, but compounds with similar functional groups are reported to possess antioxidant properties such Some derivatives containing aminocarbonitrile derivatives incorporating sugar moieties showed antioxidant activity *in vitro* (Dangolani *et al.*, 2018). In addition, dimethylamino functionalities in cyclovalone derivatives demonstrated enhanced free-radical scavenging compared to non-amine analogues, indicating that tertiary amines can contribute positively to antioxidant potency (Hayun *et al.*, 2017). There isn't much direct experimental data on this specific compound, but its chemical properties indicate it might have anticancer and antioxidant effects, which means more focused research is needed.

1,4-Butanediamine, 2,3-dimethoxy-N,N,N',N'-tetramethyl-, [S-(R*,R*)]- is a chemical compound with the molecular formula C₁₀H₂₄N₂O₂. It is chemically characterized by dimethoxy groups and tetramethylated butanediamine structure. Although no direct studies have examined 1,4-Butanediamine, 2,3-dimethoxy-N,N,N',N'-tetramethyl-, [S-(R*,R*)] for anticancer activity, analogous tetramethylated polyamine analogues have demonstrated potent cytotoxicity in human tumor cell lines (e.g., BE-Q-Amm-4-4-4 induced >90% cell death in SF-767 and SF-126 at 10 μ M) by depleting cellular polyamines (Minarini *et al.*, 2013). Additionally, symmetrically substituted polyamine derivatives triggered oxidative stress-mediated death in HeLa cells, a process attenuated by antioxidants, highlighting a potential ROS-related mechanism (Basu *et al.*, 1994).

Propane, 2-fluoro-2-methyl- (also known as 2-fluoro-2-methylpropane) is a fluorinated hydrocarbon with limited direct documentation of biological activities. Propane, 2-fluoro-2-methyl- is one of the active compounds found in Iraqi buffalo buttermilk (Al Garory and Al Musa, 2024). Although no experimental data exists on 2-fluoro-2-methylpropane itself regarding cytotoxicity, antiviral, or antioxidant activity, it has been identified (\approx 3.97%) as a minor constituent in thyme essential oil, a mixture with confirmed antimicrobial and some antioxidant effects (e.g., thymol, carvacrol) (Aljabeili *et al.*, 2018; Kuley *et al.*, 2024).

n-Hexadecanoic acid exhibits a broad range of biological actions, including anticancer, antioxidant, anti-inflammatory, antimicrobial, and hepatoprotective

properties (Dave and Ledwani, 2012; Hsu and Chung, 2012; Dong *et al.*, 2016; Sharma *et al.*, 2017; Afolayan *et al.*, 2024). It is also found as one of the active compounds of Barhi date seed ethanolic extracts (Shareef, 2025) and in Iraqi buffalo buttermilk (Al Garory and Al Musa, 2024). It has been reported to function as a natural antioxidant, scavenging free radicals and reducing oxidative stress (Afolayan *et al.*, 2024; Ganesan *et al.*, 2024). n-Hexadecanoic acid shows moderate antibacterial effects against pathogens such as *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, and *Klebsiella pneumoniae* (Ganesan *et al.*, 2024). It inhibits enzymes like cyclooxygenase-1 and -2 (COX-1 and COX-2), key inflammatory mediators (Purushothaman *et al.*, 2025). Some studies suggest n-hexadecanoic acid may inhibit cancer cell proliferation, indicating anticancer potential (Afolayan *et al.*, 2024).

Phenethyl alcohol, 2,5-dihydroxy-.alpha.-methyl-, (-)- and related phenethyl alcohol compounds have been studied for antioxidant and anticancer effects, though specific data on this exact compound is limited. Some phenethyl alcohol derivatives show antioxidant properties, helping reduce oxidative stress. For instance, studies report that phenolic derivatives of phenethyl alcohol demonstrate good antioxidant capacity (Sirilun *et al.*, 2017). While pure phenethyl alcohol itself is not strongly documented for anticancer activity, related compounds like caffeic acid phenethyl ester (CAPE), a phenylpropanoid derivative, have demonstrated tumor growth inhibition and antitumor activity (Aggarwal and Sung, 2009). CAPE stands for Caffeic Acid Phenethyl Ester. It combines caffeic acid (a natural antioxidant found in many plants) with phenethyl alcohol. CAPE is well-studied because it has several biological activities, especially anticancer, anti-inflammatory, and antioxidant effects. give exact reference (Murtaza *et al.*, 2014). Phenethyl alcohol is widely recognized for its antimicrobial properties against Gram-positive and Gram-negative bacteria, making it a common preservative and disinfectant (Corre *et al.*, 1990). Related phenethyl alcohol compounds have shown inhibition of viral DNA synthesis in some studies (Corre *et al.*, 1990).

Maltol is a naturally occurring compound widely known for its use as a flavor enhancer, Maltol also found as an active compound of milk treated with ultrasound for 5 minutes contributing to the flavor-enhancing volatile compounds (Al Musa *et al.*, 2025), but it also shows a variety of biological activities: It has demonstrated antimicrobial properties, especially when combined with cationic surfactants, increasing its activity against contaminant microorganisms (Ziklo *et al.*, 2021). Maltol (3-hydroxy-2-methyl-4-pyrone) is extensively utilized as a supplement in food and cosmetics, exhibiting antioxidant and anti-inflammatory properties. Maltol suppressed the activation of NLRP3 and NC inflammasomes, leaving other inflammasomes unaffected. Maltol also reduced IL-1 β release caused by

inflammasome activation in mice. The anti-inflammatory action of maltol was demonstrated by the decrease of reactive oxygen species (ROS) generation and Casp1 activity. Maltol is proposed to be a promising anti-inflammasome agent (Ahn *et al.*, 2022). Maltol demonstrates antioxidant and anticancer properties, enhancing its potential biological applications as well as food flavoring (Guo *et al.*, 2018).

CONCLUSIONS

This study demonstrated that *Momordica charantia* exhibits various biological activities, suggesting its potential as a natural supplement in treating numerous diseases. Furthermore, the chemical composition is highly diverse, with new bioactive compounds identified in recent years. Alternative extraction techniques, such as water or ethanol, may facilitate the identification of new secondary metabolites in this plant species.

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CONFLICT OF INTEREST

The authors affirm that there are no conflicts of interest pertaining to the publication of this manuscript.

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تحديد المواد الكيميائية النباتية النشطة بيولوجياً في المستخلص الميثانولي لفاكهة مومورديكا كارانتيا باستخدام تقنية كروماتوغرافيا الغاز-مطياف الكتلة: رؤى حول الأنشطة البيولوجية المحتملة

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الخلاصة

تناقش هذه الدراسة التركيب الحيوي النشط للمستخلص الميثانولي لثمرة المومورديكا كارانتيا. تم استخدام شرائح مجففة من ثمرة المومورديكا كارانتيا العضوية النقية 100% في تحضير المستخلص الميثانولي باستخدام جهاز الاستخلاص السوكسلت. تم إجراء التحليل الكيميائي باستخدام تقنية الكروماتوغرافيا الغازية المقترنة

بمطياف الكتلة (GC-MS)، حيث تم تحليل نواتج الفصل استنادًا إلى أطياف الكتلة الناتجة، ثم تمت مقارنتها بقاعدة بيانات الأطياف المرجعية مكتبة (NIST20.L) بهدف تحديد المركبات الكيميائية الموجودة في المستخلص. ساهمت مكونات كيميائية مختلفة في تكوين الملف الكيميائي النباتي النهائي، مما يؤثر الملفات الكيميائية النباتية التي تم تحديدها في دراسات سابقة على ثمار المومورديكا كارانتيا وشملت المركبات الرئيسية في هذه الدراسة كلاً من: 6- بنتيل بيبريدين-2-أون؛ 1،4 - حمض البنزين ثنائي الكربوكسيل؛ إستر ثنائي (2- إيثيل هكسيل)؛ مايو-إينوزيتول، 4-سي-ميثيل - 1-ميثيل-5-فلورويوراسيل؛ 1،3،5،7- تتروكسان؛ بيوتان نيتريل، 4-(ثنائي ميثيل أمينو)؛ 1،4- بيوتان دي أمين، 3،2- ثنائي ميثوكسي -ن،ن،ن،ن'- تيتراميثيل -، [س-(ر،*،ر)]-؛ بروبان، 2- فلورو-2-ميثيل -؛ ن- حمض الهكساديكانويك؛ فينيثيل كحول، 2،5 - ديهيدروكسي-ألفا-ميثيل-، (-)؛ و مالتول. وقد تم التحقق من أن هذه المركبات ومشتقاتها تتمتع بأنشطة حيوية مهمة لصحة الإنسان، مثل: التأثيرات المضادة للبكتيريا، والمضادة للسرطان، والمضادة للأكسدة، والمضادة للالتهاب، والمضادة للميكروبات، والحامية للكبد، والمضادة للفيروسات، والمضادة للملاريا، وخافضة للحرارة، والمضادة للأورام. تُعد هذه التأثيرات البيولوجية لمستخلص ثمار المومورديكا كارانتيا الميثانولي ذات صلة مباشرة بالصحة العامة، وتُبرز نتائج هذه الدراسة أهميتها في توجيه البحوث المستقبلية نحو استخلاص المركبات الفعالة بيولوجيًا من هذه الثمرة وتطبيقاتها الطبية الممكنة.

الكلمات المفتاحية: القرع المر، الثمرة، التفاح المر، القرع المر، التأثيرات الحيوية.

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