

GC-MS PROFILING OF BIOACTIVE COMPOUNDS AND ASSESSMENT OF POLYPHENOLIC CONTENT AND ANTIOXIDANT ACTIVITY IN *CUMINUM CYMINUM* SEEDS

MD NAUAZ SHARIF FAHAD¹, MEHEDI MAHMUDUL HASAN², YEASIN ARAFAT¹,
MARIA AHMED CHANDRA¹, MD MAZBA UDDIN CHOWDHURY¹, MANTI SAHA¹
AND MOHAMMAD ASADUZZAMAN^{1*}

Department of Biochemistry and Molecular Biology, Noakhali Science and Technology University, Noakhali-3814, Bangladesh

Keywords: Cumin, Antioxidant, Phytochemical, Radical, GC-MS, Bioactive

Abstract

This study investigates the phytochemical composition, antioxidant potential, and chemical profile of cumin seeds (*Cuminum cyminum*) using phytochemicals (polyphenols, flavonoids and tannins) screening, DPPH antioxidant assay, and Gas Chromatography–Mass Spectrometry (GC–MS) analysis. Results revealed significant levels of polyphenols (36.42%), flavonoids (32.86%), and tannins (17.41%) in the ethanolic extract, which also showed the highest extraction yield (10.61%) compared to the aqueous extract. Antioxidant activity, assessed via DPPH assay, showed stronger radical scavenging in ethanolic extract (IC₅₀ = 101.95 µg/ml) than water extract (IC₅₀ = 147.55 µg/ml). GC-MS analysis identified various fifty bioactive compounds in the extracts. These findings highlight cumin seeds as a rich source of phenolics with notable antioxidant properties, supporting their potential use in functional foods and natural therapeutics.

Introduction

Cuminum cyminum L., commonly known as cumin, is a flowering plant species belonging to Apiaceae (Pandey *et al.* 2022). Cumin is widely known as 'jeera' in Bangladesh. Cumin seed, known for its aroma and antioxidant power, is gaining importance in Bangladesh. Its health benefits may help to reduce costs from oxidative stress-related diseases, while its use in medicine and diet underscores its wide-ranging value. Cumin reduces reactive oxygen species (ROS) with its strong antioxidants, protecting DNA, proteins, and lipids from damage and lowering the risk of chronic diseases (Siger *et al.* 2008). Cumin, like many plants, is rich in antioxidants (Fatima *et al.* 2018) and anti-aging compounds, mainly due to its high phenolic acid content and other bioactive that help to protect against oxidative stress (Li and Jiang 2004). To fight oxidative stress, the body uses its own enzymes (SOD, CAT, GPx) and dietary antioxidants (Pizzino *et al.* 2017). Natural sources like cumin offer both nutritional support and disease prevention, highlighting the value of diet in promoting health (Babu *et al.* 2013).

GC-MS combines gas chromatography and mass spectrometry to separate and precisely identify bioactive compounds in cumin seeds. It effectively analyzes essential oils, phenolic acids, and other volatile components, providing insights into cumin's antioxidant potential. This method offers high sensitivity and precision, aiding the identification of key antioxidants and bioactive compounds with potential health benefits (Adams 2017).

This research aims to explore cumin seeds antioxidant properties and their potential for promoting health and reducing oxidative stress. By analyzing its bioactive compounds through advanced extraction, phytochemical analysis, and Gas Chromatography-Mass Spectrometry (GC-MS) profiling, the study seeks to enhance its medical and dietary applications. It addresses the gap

*Author for correspondence: <asad.bmb@nstu.edu.bd>. ¹Department of Biochemistry and Molecular Biology, Noakhali Science and Technology University, Noakhali-3814, Bangladesh. ²Department of Fisheries and Marine Science, Noakhali Science and Technology University, Noakhali-3814, Bangladesh.

in scientific knowledge about cumin seeds composition and antioxidant effects, particularly in Noakhali, and aims to highlight their therapeutic potential.

Materials and Methods

The experiment was conducted at the Laboratory of the Department of Biochemistry and Molecular Biology, Noakhali Science and Technology. Cumin seeds were collected from the local market of Noakhali. The seeds were thoroughly washed with distilled water and then air-dried. The dried seeds were ground into a fine powder using a mechanical grinder to facilitate extraction. For the preparation of extracts, 20 g of cumin seeds powder was used for each treatment. Two solvent systems were employed: 100 ml 70% ethanol for the ethanolic extraction and 100 ml of distilled water for the aqueous extraction. Powder and solvent mixtures were placed in clean, amber-colored glass containers to prevent light-induced degradation and were shaken intermittently for 72 hrs in a dark environment. The mixtures were filtered through Whatman No. 1 filter paper. The filtrates were concentrated using a rotary evaporator at 40°C for ethanol extracts and 55°C for aqueous extracts under reduced pressure. The resulting dried extracts were stored at -20°C in sterile, airtight vials until further analysis.

Phytochemical analyses were conducted to determine the total polyphenol, flavonoid, and tannin contents of the extracts. The total polyphenol content (TPC) was measured using the Folin–Ciocalteu reagent, following the method of Singleton *et al.* (1999) as modified by Afroz *et al.* (2014). The total flavonoid content (TFC) was estimated via the aluminum chloride colorimetric method developed by Chang *et al.* (2002). The total tannin content (TTC) was determined using the Folin–Ciocalteu method as described by Tambe and Bhambar (2014).

To assess antioxidant activity, the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay was performed. The method followed was based on the protocol of Braca *et al.* (2002), with absorbance measured spectrophotometrically. The percentage of DPPH radical scavenging activity was calculated for each extract to evaluate their antioxidant potential. The chemical composition of the 70% ethanolic cumin seed extract was analyzed using a GC-MS system.

Results and Discussion

The initial cumin seed dry powder weight used for extraction was 109.67 g for the ethanolic extract and 104.37 g for the water extract. The extract yield obtained was 11.6 g from the ethanolic extract and 9.3 g from the water extract. This corresponded to a percentage yield of 10.61% for the ethanolic extract and 8.91% for the water extract.

As for polyphenol, the total content of cumin seeds ethanolic extract was found to be 36.42 ± 0.003 g GAE/100 g sample and water extract showed the value up to 28.72 ± 0.009 g GAE/100 g of sample. Polyphenols expressed as Gallic Acid Equivalent, represent a major class of antioxidant compounds in plant materials. The superior polyphenol levels in the ethanolic extract align with ethanol's effectiveness in extracting less polar phenolic compounds, contributing to the observed stronger antioxidant activity in this extract. These findings align with previous studies (Afroz *et al.* 2014, Denre 2014), where ethanol was noted to extract more polyphenolic compounds due to its higher polarity (Do *et al.* 2014).

The water extract demonstrated a markedly higher flavonoid content of 32.86 ± 0.031 g Catechin Equivalent (CE) per 100 g of sample, while the ethanolic extract showed 12.02 ± 0.005 g CE/100 g of sample. Flavonoids, quantified and expressed as Catechin Equivalent, are a subclass of polyphenols known for their potent free radical scavenging properties and metal-chelating abilities. These flavonoids play a critical role in reducing oxidative stress by neutralizing reactive

oxygen and nitrogen species (Tilak *et al.* 2004). Despite the lower yield in ethanol, cumin remains a rich source of flavonoids with potential therapeutic benefits.

Tannin content was higher in the water extract (17.41 ± 0.013 g TAE/100 g) than in the ethanolic extract (11.93 ± 0.014 g TAE/100 g). Tannins, quantified as Tannic Acid Equivalent (TAE), are polyphenolic compounds that bind proteins and metals, contributing to astringency and plant antioxidant defense.

Antioxidant activity of the extracts was determined by DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay and expressed as half maximal inhibitory concentration (IC_{50} , $\mu\text{g/ml}$), which were calculated from a plot inhibition percentage vs. extract concentration. The ethanolic extract exhibits more potent free radical scavenging activity with IC_{50} value of $101.95 \mu\text{g/ml}$. On the other hand, water extracts yields a higher IC_{50} of $147.55 \mu\text{g/ml}$ which indicates a lesser antioxidant activity in scavenging DPPH radicals (Fig. 1). The ethanolic extract had higher total polyphenol content, and may have stronger antioxidant capability than aqueous extracts.

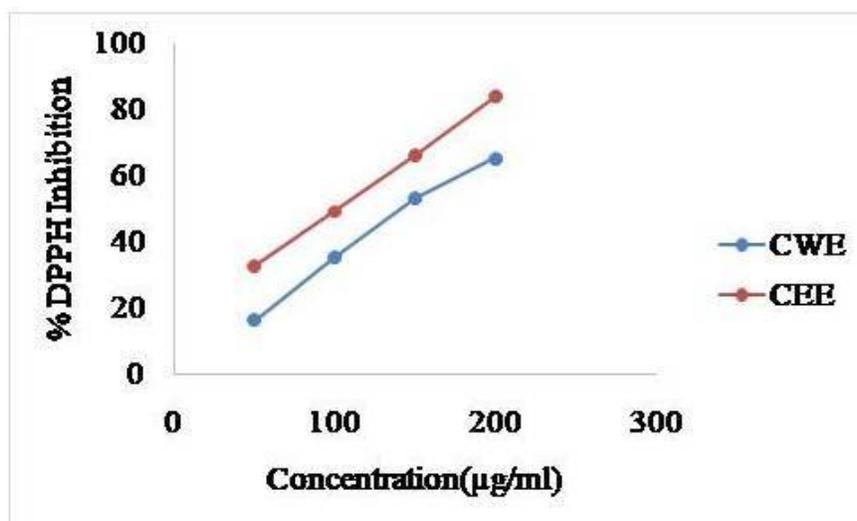


Fig. 1. DPPH radical scavenging activity of ethanolic (CEE) and aqueous (CWE) extracts of cumin seeds at various concentrations.

Gas chromatography-mass spectrometry (GC-MS) analysis revealed a wide spectrum of bioactive compounds in cumin seeds (Table 1). Ethanol, 2-(2-butoxyethoxy) and dimethyl sulfoxide were detected as surface-active agents with antimicrobial potential. pDioxane-2,3-diol was associated with enzyme inhibition, cytotoxicity, and antioxidant activity. Phenol, although found in low concentration, is recognized for its disinfectant action. Compounds such as 2-propen-1-ol and sulfurous acid, cyclohexylmethyl hexadecyl ester, also supports the antimicrobial effects of cumin seeds.

Additional antimicrobial constituents included 2-ethyl-5-n-propylphenol, benzyl o-nitrobenzoate, and n-pentadecanol, the latter known for its broader biological roles including anticancer and insecticidal activities. Sugar derivatives such as methyl- β -D-thiogalactoside and β -D-glucopyranoside contributed enzyme-inhibiting, antioxidant, and antimicrobial properties. 4-ethyl-3-oxabicyclo[4.4.0]decane also play role in antimicrobial and cytotoxic functions.

Table 1. Bioactive compounds detected by GC-MS in cumin seed extracts along with molecular formulas.

Sl. No.	Bioactive compounds	Molecular formula
1	1,2,4-Butanetriol	C ₄ H ₁₀ O ₃
2	1,2-Cyclopentanedione	C ₅ H ₆ O ₂
3	1,4-Cyclohexadiene-1-methanol, 4-(1-methylethyl)-acetate	C ₁₂ H ₁₈ O ₂
4	13-Docosenamide	C ₂₂ H ₄₃ NO
5	1-Pentadecene	C ₁₅ H ₃₀
6	2-(Isobutoxymethyl)oxirane	C ₈ H ₁₆ O ₂
7	2,3,5-Trimethylanisole	C ₁₀ H ₁₄ O
8	2-Ethyl-5-n-propylphenol	C ₁₁ H ₁₆ O
9	2-Methoxy-4-vinylphenol	C ₉ H ₁₀ O ₂
10	2-Oxabicyclo[2.2.2]octan-6-ol, 1,3,3-trimethyl-	C ₁₀ H ₁₈ O ₂
11	2-Propen-1-ol (Allyl alcohol)	C ₃ H ₆ O
12	3,5-Heptadienal, 2-ethylidene-6-methyl-	C ₁₀ H ₁₄ O
13	3-Hexadecanol	C ₁₆ H ₃₄ O
14	3-Tetradecene	C ₁₄ H ₂₈
15	4-Ethyl-3-oxabicyclo[4.4.0]decane	C ₁₂ H ₂₀ O
16	4-Isopropylcyclohexa-1,3-dienecarbaldehyde	C ₁₀ H ₁₂ O
17	6-Octadecenoic acid	C ₁₈ H ₃₄ O ₂
18	7-Hexadecenal (Z)-	C ₁₆ H ₃₀ O
19	8-Hydroxycarvotanacetone	C ₁₀ H ₁₆ O ₂
20	8-Methyl-nonenamide	C ₁₀ H ₁₉ NO
21	Acetic acid, chloro-, hexadecyl ester	C ₁₈ H ₃₅ ClO ₂
22	aR-Turmerone	C ₁₅ H ₂₂ O
23	Benzaldehyde, 4-(1-methylethyl)- (p-Isopropylbenzaldehyde)	C ₁₀ H ₁₂ O
24	Benzenepropanal, β-methyl	C ₁₀ H ₁₂ O
25	Benzyl o-nitrobenzoate	C ₁₄ H ₁₁ NO ₄
26	E,E,Z-1,3,12-Nonadecatriene-5,14-diol	C ₁₉ H ₃₄ O ₂
27	D-Allose	C ₆ H ₁₂ O ₆
28	Dimethyl sulfoxide (DMSO)	C ₂ H ₆ OS
29	Dotriacontane	C ₃₂ H ₆₆
30	Ethanol, 2-(2-butoxyethoxy)	C ₈ H ₁₈ O ₃
31	Ethanol, 2-[2-(2-methoxyethoxy)ethoxy]-	C ₇ H ₁₆ O ₄
32	Ethyl 13-methyl-tetradecanoate	C ₁₇ H ₃₄ O ₂
33	Ethylene glycol, TMS derivative	C ₅ H ₁₄ O ₂ Si
34	Ethyl-α-D-glucopyranoside	C ₈ H ₁₆ O ₆
35	Linoleic acid ethyl ester	C ₂₀ H ₃₆ O ₂
36	Methyl 10-trans,12-cis-octadecadienoate	C ₁₉ H ₃₄ O ₂
37	Methyl-β-D-thiogalactoside	C ₇ H ₁₄ O ₅ S
38	N,N-Dimethylaminoethanol	C ₄ H ₁₁ NO
39	n-Pentadecanol	C ₁₅ H ₃₂ O
40	Octadecane, 5-methyl-	C ₁₉ H ₄₀
41	o-Ethylhydroxylamine	C ₂ H ₇ NO
42	Oxalic acid, cyclohexylmethyl tetradecyl ester	C ₂₇ H ₄₈ O ₄
43	p-Cymen-7-ol	C ₁₀ H ₁₄ O
44	p-Dioxane-2,3-diol	C ₄ H ₈ O ₄
45	Phenol	C ₆ H ₆ O
46	p-Menth-2-en-7-ol	C ₁₀ H ₁₈ O
47	Spiro[4.4]nona-1,3-diene, 1,2-dimethyl	C ₁₁ H ₁₆
48	Sulfurous acid, cyclohexylmethyl hexadecyl ester	C ₂₃ H ₄₆ O ₃ S
49	β-D-Glucopyranose, 4-O-β-D-galactopyranosyl	C ₁₂ H ₂₂ O ₁₁
50	β-D-Glucopyranoside, methyl	C ₇ H ₁₄ O ₆

Several antioxidant compounds were also identified. O-ethylhydroxylamine, 1,2-cyclopentanedione, and 1,2,4-butanetriol contributed to radical scavenging and system stabilization. p-Menth-2-en-7-ol and 2-methoxy-4-vinylphenol exhibited multiple functions including antimicrobial and anti-inflammatory activity. Sugar-based antioxidants such as D-allose, ethyl α -D-glucopyranoside, and β -D-glucopyranose show strong biological activity, while α R-turmerone and linoleic acid ethyl ester add neuroprotective and cardiovascular benefits.

Anti-inflammatory compounds were also prominent. Ethyl α -D-glucopyranoside, α R-turmerone, and p-menth-2-en-7-ol showed consistent immune-modulating effects. Fatty acid esters like ethyl 13-methyl-tetradecanoate and methyl 10-trans,12-cis-octadecadienoate demonstrated anti-inflammatory and antioxidant activity. Amide derivatives such as 13-docosenamide and 8-methyl-nonenamide contribute to anti-inflammatory and mild analgesic properties.

This study shows that cumin seeds from Noakhali are rich in antioxidants, particularly when extracted with ethanol. The presence of polyphenols, flavonoids, and tannins, along with bioactive compounds identified by GC-MS, highlights cumin's health and industrial potential. However, further in vivo studies and clinical trials are needed to confirm therapeutic effects, and optimizing extraction methods.

Acknowledgement

Thanks to the Biochemistry and Molecular Biology Laboratory team for their invaluable resources and guidance.

References

- Adams RP 2017. Identification of essential oil components by gas chromatography/mass spectrometry (5 online Ed). Gruver, Texensis Publishing, TX USA.
- Afroz R, Tanvir E, Islam MA, Alam F, Gan SH and Khalil MI 2014. Potential antioxidant and antibacterial properties of a popular jujube fruit: Apple Kul (*Zizyphus mauritiana*). J. Food Biochem. **38**(6): 592-601.
- Babu PVA, Liu D and Gilbert ER 2013. Recent advances in understanding the anti-diabetic actions of dietary flavonoids. J. Nutr. Biochem. **24**(11): 1777-1789.
- Braca A, Sortino C, Politi M, Morelli I and Mendez J 2002. Antioxidant activity of flavonoids from *Licania licaniaeflora*. J. Ethnopharmacol. **79**(3): 379-381.
- Chang CC, Yang MH, Wen HM and Chern JC 2002. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. J. Food Drug Anal. **10**(3): 178-182.
- Denre M 2014. The determination of vitamin C, total phenol and antioxidant activity of some commonly cooking spices crops used in West Bengal. Int. J. Plant Physiol. Biochem. **6**(6): 66-70.
- Do QD, Angkawijaya AE, Tran-Nguyen PL, Huynh LH, Soetaredjo FE, Ismadji S and Ju YH 2014. Effect of extraction solvent on total phenol content, total flavonoid content, and antioxidant activity of *Limnophila aromatica*. J. Food Drug Anal. **22**(3): 296-302.
- Fatima T, Beenish NB, Gani G, Qadri T and Bhat TA 2018. Antioxidant potential and health benefits of cumin. J. Med. Plants Stud. **6**: 232-236.
- Li R and Jiang ZT 2004. Chemical composition of the essential oil of *Cuminum cyminum* L. from China. Flav. Fragr. J. **19**(4): 311-313.
- Pandey P, Awasthi R, Dhiman N, Sharma B and Kulkarni GT 2022. Ethnopharmacological reports on herbs used in the management of tuberculosis. In M. Sarwat & H. Siddique (Eds), Academic Press, USA. pp. 501-523.
- Pizzino G, Irrera N, Cucinotta M, Pallio G, Mannino F, Arcoraci V, Squadrito F, Altavilla D and Bitto A 2017. Oxidative stress: harms and benefits for human health. Oxid. Med. Cell. Longev. **2017**(1): 8416763.

- Siger A, Nogala-Kalucka M and Lampart-Szczapa E 2008. The content and antioxidant activity of phenolic compounds in cold-pressed plant oils. *J. Food Lipids* **15**(2): 137-149.
- Singleton VL, Orthofer R and Lamuela-Raventós RM 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. *Methods Enzymol.* **299**: 152-178.
- Tambe VD and Bhambar RS 2014. Estimation of total phenol, tannin, alkaloid and flavonoid in *Hibiscus tiliaceus* Linn. wood extracts. *J. Pharmacogn. Phytochem.* **2**(4): 41-47.
- Tilak JC, Banerjee M, Mohan H and Devasagayam T 2004. Antioxidant availability of turmeric in relation to its medicinal and culinary uses. *Phytother. Res.* **18**(10): 798-804.

(Manuscript received on 15 May, 2025; revised on 29 November, 2025)