J. Bangladesh Acad. Sci. 49(2); 243-253: December 2025 DOI:https://doi.org/10.3329/jbas.v49i2.80644

Journal of Bangladesh Academy of Sciences

Journal homepage: http://www.bas.org.bd/publications/jbas.html

Research Article

Phytochemical and antibacterial properties of clove from India, Sri Lanka, and Indonesia available in Bangladesh

Sharmin Hossen, Irin Hossain[1], Shamim Akhtar[1], Arpita Chowdhury[*],
Mohammed Tanvir Ahamed, Susmita Chowdhury and Monsur Ahmad

Department of Applied Chemistry & Chemical Technology, Faculty of Food Science and Technology
Chattogram Veterinary and Animal Sciences University, Chittagong, Bangladesh

ARTICLE INFO

Article History

Received: 19 March 2025
Revised: 19 May 2025
Accepted: 25 August 2025

Keywords: *Syzygium aromaticum*, Clove, Essential oil, Phytocomponent, GC-MS profiling, Antibacterial activity, Antioxidant activity.

ABSTRACT

Cloves (*Syzygium aromaticum*) are aromatic flower buds that have been extensively studied for their culinary, medicinal, and economic value. Bangladesh has Indonesian (CIS), Indian (CID) and Sri Lankan (CSI) cloves. This study investigates the bioactive compounds, proximate analysis, physicochemical properties, and antibacterial efficacy of clove essential oils from three commercially available brands in Chattogram. Using Gas Chromatography-Mass Spectrometry (GC-MS), the primary bioactive compounds identified were eugenol, eugenol acetate, and β-caryophyllene, with significant variations in their concentrations across brands. Proximate analysis revealed differences in moisture, ash, and volatile content, highlighting disparities in quality and purity. Physicochemical properties were assessed. Antibacterial efficacy was evaluated using the agar disc diffusion method against *Escherichia coli* and *Staphylococcus aureus* and *Pseudomonas aeruginosa*, as indicated by its minimal inhibitory concentration (MIC) values. These findings underscore the importance of brand selection in ensuring the therapeutic and functional efficacy of clove essential oil, emphasizing the need for stringent quality control in the market.

Introduction

Cloves (*Syzygium aromaticum*) are the fragrant desiccated flower buds of a tree belonging to the Myrtaceae family (Srivastava and Malhotra, 1991). Exotic goods imported from Asia were particularly appealing to the Greeks and Romans. The Portuguese, Dutch, Spanish, and British were driven to expand into countries such as India, Sri Lanka, and Indonesia by their ambition to control sources of spices. Every country fought with others to gain monopolistic control over main trade routes and areas of spice production (Takeda et al., 2008). For the reason that cloves from all these three locations are abundant in essential oils, predominantly comprising eugenol. Eugenol, which comprises 72-90% of the essential oil derived from cloves, is responsible for their distinctive fragrance and numerous biological functions (Alma et al., 2007). Clove essential oil (CEO) extracted from all parts of clove tree are a rich source of bioactive compounds with potential antioxidant, antimicrobial and

[*]Corresponding author:<arpitachy23@gmail.com>

[1]*Department of Chemistry, University of Chittagong, Chittagong, Bangladesh;*

[*]*Department of Food Processing and Engineering, Chattogram Veterinary and Animal Sciences University, Bangladesh*

anti-inflammatory properties. The CEO also combats dental pain and oxidative stress. Principal health advantages comprise: antioxidant properties (Jirovetz et al., 2006), antibacterial and antifungal effects (Nzeako et al., 2006), blood sugar regulation (Mohan et al., 2019), cancer prevention (Liu et al., 2014). Researchers have been investigating the potential health advantages of clove oil and its constituents, similar to other EOs (Haro-González et al., 2021). In a prior study, researchers identified the components of EO derived from the leaves and buds (Bhuiyan et al., 2010). Cloves, particularly their bioactive compounds like eugenol, may offer natural alternatives to synthetic antibiotics, against *E. coli*, *Staph. aureus*, and *P. aeruginosa*, helping combat antibiotic resistance (Jayarathne et al., 2020). As these bacteria are often resistant to conventional antibiotics, cloves could provide a natural solution, potentially reducing reliance on synthetic drugs and slowing the development of resistance. To summarize, this study has the potential to provide natural remedies for health problems, make food more secure, and bolster long-term health and food industry sustainability of the food industry.

Materials and methods

Collection and processing of cloves

Cloves were procured from the Khatunganj market in Chittagong City. The samples were purified, dust particles removed, and dried. The dried cloves were ultimately pulverized using a Fritsch mortar grinder (Germany) for 1 hour.

Extraction of Essential Oil (EO)

Plant essential oil extraction uses several methods. In this investigation, steam distillation using the Clevenger apparatus (Germany) was performed according to the steps described by Shavisi et al. (2017), with slight moderations.

Determination of physicochemical properties of EO

The physical parameters of the extracted EO, including refractive index, density, optical rotation, and alcohol solubility, were assessed following established methods (Pharmacopoeia Commission, 2011) with three replications.

Determination of acid value

The Acid value was estimated using the following equation mentioned in Paez et al. (2016):

$$\text{Acid value} = \frac{\text{N of alkali (0.1) x ml of alkali}}{\text{Wt. of sample (gm)}}$$

Determination of saponification value

The saponification value was estimated using the following equation mentioned in Paez et al. (2016):

$$\text{Saponification value} = \frac{\text{N x (b-a) x 56.1}}{\text{Wt. of sample (gm)}}$$

Where, b = blank titer value, a = sample titer value and N = 0.5 (Normality) of HCl.

Determination of peroxide value

Peroxide value was determined using the formula described by Paez et al. (2016) incorporating $Na_2S_2O_3$.

Determination of Iodine value

Iodine value was determined using methods mentioned in Paez et al. (2016).

Identification of unidentified chemicals in clove oil via GC-MS analysis

The analysis was conducted using GC-MS electron impact ionization (EI) on a GC-17A gas chromatograph (Shimadzu, Japan) coupled to a GC/MS-QP 5050A mass spectrometer (Shimadzu). The column temperature was programmed from 40°C (for 2 minutes) to 250°C at a rate of 5°C/min (Aziz et al., 2012).

Extract Preparation

Making of an Extract by adding 10 ml of 100% ethanol + acetone to 1g of material in a Falcon tube and then letting it sit for 72 hours. After 72 hours, filtered the solvent and collected the filtrates. Then, the extract was analyzed.

Determination of antioxidant activity (AOA) by DPPH scavenging method

Antioxidant mobility of the extracts was determined using DPPH assay described by Akther et al. (2023) with slight modifications. The absorbance was measured at 517 nm using a UV-VIS spectrophotometer (UV-2600, Shimadzu Corporation, USA).

Determination of Bioactive Compounds

Extract Preparation

The extraction process involves combining 10 ml of pure ethanol (100%) with 1 g of the substance in a Falcon tube, then allowing it to rest undisturbed for a duration of 72 hours. After 72 hours, strain the solvent and collect the resulting filtrates. Subsequently, the ethanoic extract was identified.

Total Flavonoid Content (TFC)

The total flavonoids content (TFC) of the clove oil samples was determined using the aluminum chloride colorimetric method reported by Chang et al. (2002) with slight modifications. A suitable volume of extract stock solution (1 mg/mL) was prepared for the calibration curve. Quercetin (Sigma, USA) was dissolved in 80% ethanol to generate standard solutions at concentrations of 0.025, 0.050, 0.075, and 0.100 mg/ml. In the cuvette, 0.5 mL aliquots of the standard solution (diluted extract) were mixed with 1.5 mL of 95% ethanol, 0.1 mL of 10% aluminium chloride (QualiChem, India), 0.1 mL of 1 mol/L potassium acetate (Merck, Germany), and 2.8 mL of distilled water. The obtained mixture was allowed to cool to room temperature (25.5 ± 1°C) for 30 minutes prior to use. The absorbance at 415 nm was subsequently measured via a UV-visible spectrophotometer. In the blank preparation, 10% aluminium chloride was substituted with an equivalent volume of distilled water. Total flavonoid content (TFC) was quantified in milligrammes of quercetin equivalents (QE) per 100 grammes of extract.

Measurement of Polyphenols (TPC)

The total polyphenol content (TPC) of the CEO was determined using the Folin-Ciocalteu method reported by Al-Owaisi et al. (2014) with slight modifications. Appropriate stock solutions (1 mg/mL) of extracts and standard solutions (0.02, 0.04, 0.06, 0.08, and 0.10 mg/mL) of gallic acid (Sigma, USA) were formulated for the experiment. Subsequently, 0.3 mL of gallic acid standard solution or extracts was pipetted into a cuvette. Subsequently, 1.5 mL of the diluted Folin-Ciocalteu reagent was added, and the mixture was mixed. After a 3-minute pause, 1.5 mL of sodium carbonate solution (75 g/L) was added, and the mixture was allowed to stand for 60 minutes. Ethanol served as the blank for absorbance measurement at 765 nm utilising a UV spectrophotometer. Total phenolic content (TPC) was quantified in milligrammes of gallic acid equivalents (GAE) per 100 grammes of extracts.

Quantification of Anthocyanins (TAC)

Total anthocyanin content (TAC) of clove oil extracts was determined colorimetrically following the process expressed with slight modifications (Akther et al., 2023). A precise 10 mg/mL stock solution of extracts was formulated for the experiment. Furthermore, 3 mL aliquots of the extract solution were transferred into a cuvette. The colour concentration was measured at 520 nm using a UV-vis spectrophotometer (UV-2600, Shimadzu). Ethanol (Merck, Germany) served as the control sample. TAC was measured in mg/100 g with the subsequent equation:

TAC = (Absorbance of sample) x (DF) x (100) / (m) x (E)

Where, DF denotes the dilution factor, m signifies the weight of the sample utilised to make a stock solution, and E indicates the extinction coefficient, valued at 55.9.

Determination of antibacterial activity of clove oil

The antibacterial activity was evaluated using the disc diffusion method, commonly referred to as the Kirby-Bauer test, as stated in Drew et al. (1972). Sterile saline silution was used as nagative control.

Statistical Analysis

The obtained data were stored in Microsoft Excel 2013 spreadsheet to evaluate statistical analysis, and significant differences were determined using one-way analysis of variance (ANOVA). All samples were in three replicates. Data were sorted, coded and recorded in IBM SPSS Statistics 25. The statistical analysis was conducted for at 5% level of significant ($p \leq 0.05$).

Results and Discussions

Physicochemical properties of clove essential oil

Table 1 and 2 represent the physicochemical properties of CEO, which were found to be as follows: the acid value, saponification value, peroxide value, Iodine value, Density and Viscosity.

The acid value (AV) is an important determinant of oil quality (Gharby et al., 2017). The acid values were determined to be 5.213 mg KOH/g oil, 6.09 mg KOH/g oil, and 6.457 mg KOH/g oil for the CIS, CID, and CSI clove essential oils, respectively. In this investigation, the acid values of all clove essential oil samples exceeded the maximum limit. The principal mechanism resulting in rancidity is lipid oxidation. Heat can expedite this process, as seen by essential oils such as nutmeg, which exhibit enhanced free radical-scavenging activity, leading to the production of aldehydes and other chemicals that promote rancidity and alterations in chemical composition upon heating (Tomaino et al., 2005).

The saponification values (SV) were determined to be 37.257 mg/g oil, 40.670 mg/g oil, and 38.056 mg/g oil with regard to CIS, CID, and CSI clove essential oils, respectively. This value closely aligned with those reported by numerous studies (Alanazi et al., 2022; Sulieman et al., 2007). The Peroxide value was found to be 4.61, 6.667 and 5.127 meq O_2/kg oil of the CIS, CID and CSI clove essential oil respectively. The peroxide value serves as an indicator of the stability and quality of oils and fats (Zahir et al., 2017). In this investigation, the PV of clove essential oil samples from CIS was lower than that of the other two samples. The unsaturation of fats and oils is shown by the iodine value. Higher unsaturated fats and oils have higher iodine values.

For the oil sample used in this investigation, the iodine value was 51.457 g I_2/100 g of oil for CIS, 49.507 g I_2/100 g of oil for CID, and 50.157 g I_2/100 g of oil for CSI clove essential oil, respectively. Kyriakidis and Katsiloulis (2000) state that the recommended range for semi-drying oils is between 100 and 300, which is far lower than this amount. The iodine value (IV) of CIS clove essential oil samples was lower than that of the other two samples in this study.

The density (ρ) and viscosity (η) of three brands of clove essential oil were determined at 6 distinct temperatures ranging from 25°C to 50°C, with 5°C intervals. All density (ρ) results are presented in Table 2. It can be observed regarding density that at a specific temperature, the density (ρ) of clove essential oil falls in the following order: CID > CSI > CIS. The ρ vs T comparison of 3 brands of clove essential oil exhibits a comparable tendency, with p falling nearly linearly as temperature increases. This result is in accordance with Porter and Lammerink (1994). As the temperature rises, the kinetic energy of the molecules in the essential oil increases, leading to their separation. This expansion results in reduced density (Gandova et al., 2024).

The subsequent observations regarding viscosity (η) are - at a specific temperature, the viscosity (η) of clove essential oil diminishes in the following sequence: CSI > CID > CIS. The η vs T comparison for 3 brands of clove essential oil exhibits a comparable tendency, with η gradually dropping as the temperature rises. This results from increased heat energy, which diminishes the intermolecular interactions within the oil, allowing the molecules to move more freely. At elevated temperatures, the kinetic energy of the molecules

in the essential oil escalates. This energy exceeds the intermolecular forces binding the molecules, resulting a reduced viscosity (Gandova et al., 2024).

Phytocomponents identified in clove bud essential oils by GC-MS analysis

Table 3 presents the phytocomponents identified in CEOs, primarily Aniline, Eugenol, Phenol, Caryophyllene oxide etc. CEO demonstrates significant antioxidant and antibacterial properties, principally due to its Eugenol content. Therefore, this oil is widely used in the food industry as a preservative and flavoring agent due to its antibacterial and antioxidant characteristics. It is employed in cosmetics for its aroma and medicinal properties (Liñán-Atero et al., 2024). Eugenyl acetate, another important ingredient, adds to the oil's scent and possible health advantages

Table 1. Physicochemical properties of clove essential oil.

Country of origin	Acid value (mg KOH /g oil)	Saponification value (mg /g oil)	Peroxide value (meq O_2/kg oil)	Iodine value (g I_2/100 g of oil)
CIS	5.213±0.01^{c}	37.257±0.01^{c}	4.610±0.01^{c}	51.457±0.01^{a}
CID	6.087±0.01^{b}	40.670±0.01^{a}	6.667±0.01^{a}	49.507±0.01^{c}
CSI	6.457±0.01^{a}	38.056±0.01^{b}	5.127±0.01^{b}	50.157±0.01^{b}

Mean ± S.D. for three replicates (n=3) and different superseripts indicate differences among orgin of clove. CIS = Indonesian origin; CID = Indian origin; CSI = Sri Lanka origin

Table 2. Density and viscosity of clove essential oil.

Temperature (^{o}C)	Density (ρ)			Viscosity (η)		
	CIS	CID	CSI	CIS	CID	CSI
25	1.02±0.005^{c}	1.03±0.005^{a}	1.03±0.005^{ab}	10.32±0.005^{c}	14.43±0.005^{b}	16.35±0.005^{a}
30	1.01±0.005^{c}	1.03±0.005^{a}	1.02±0.00^{b}	8.36±0.005^{c}	11.34±0.005^{b}	12.94±0.005^{a}
35	1.01±0.005^{c}	1.02±0.005^{a}	1.02±0.005^{ab}	6.66±0.005^{c}	9.12±0.001^{b}	10.53±0.005^{a}
40	1.00±0.005^{c}	1.02±0.005^{a}	1.02±0.005^{ab}	5.63±0.005^{c}	7.56±0.005^{b}	8.67±0.005^{a}
45	1.00±0.005^{c}	1.01±0.005^{a}	1.01±0.005^{ab}	4.82±0.005^{c}	6.36±0.005^{b}	7.31±0.005^{a}
50	0.99±0.00^{c}	1.01±0.005^{a}	1.01±0.057^{ab}	4.17±0.005^{c}	5.44±0.005^{b}	6.26±0.005^{a}

Mean ± S.D. for three replicates (n=3) and different superseripts indicate differences among orgin of clove. CIS = Indonesian origin; CID = Indian origin; CSI = Sri Lanka origin

Table 3. Phytocomponent commponents identified in clove essential oils by GC-MS analysis.

Country of origin		Compounds name	Chemical formula	MW	Retention Index (RI)
CIS	1.	2,5-Diethylideneoctahydropentalene	$C_{12}H_{18}$	162	1229
	2.	3,5-Methanocyclopentapyrazole, 3,3a,4,5,6,6a-hexahydro-3a,4,4-trimethyl-	$C_{10}H_{16}N_2$	164	0
	3.	P-Picoline (Methylpyridine)	C_6H_7N	93	787
	4.	Alpha-picoline (Methylpyridine)	C_6H_7N	93	787
	5.	Aniline (Phenylamine)	C_6H_7N	93	992
	6.	Chavicol (Hydroxyallylbenzene)	$C_9H_{10}O$	134	1203
	7.	Eugenol (2-methoxy-4-(2-propenyl)	$C_{10}H_{12}O_2$	164	1392
			$C_{12}H_{14}O_3$	206	1552
CID	8.	Trans-Z-alpha. Bisabolene epoxide	$C_{15}H_{24}O$	220	1531
	9.	Spiro[androst- 5-ene-17,1cyclobutan] -2'-one,3-hydroxy-,(3β, 17β)-	$C_{22}H_{32}O_2$	328	2413
	10.	Caryophyllene oxide	$C_{15}H_{24}O$	220	1507
	11.	1, 7, 7-Trimethylbicyclo [2.2.l] heptane-2, 5-diol	$C_{10}H_{18}O_2$	170	1326
	12.	Eugenol (2-methoxy-4-(2-propenyl)	$C_{10}H_{12}O_2$	164	1392
	13.	Phenol, 2-rnethoxy-3-(2-propenyl)-	$C_{10}H_{12}O_2$	164	1392
	14.	Phenol, 2-Allyl-4-propenyl	$C_{10}H_{12}O_2$	164	1392
	15.	Ylangene	$C_{15}H_{24}$	204	1221
	16.	Eugenyl acetate	$C_{12}H_{14}O_3$	206	1552
CSI	17.	Ethanone,1-(2,3,4-Trimethoxyphenyl)	$C_{11}H_{14}O_4$	210	1596
	18.	4-(2,6,6-Trimethyl-1-cyclohexen-l-yl) butanoic acid	$C_{13}H_{22}O_2$	210	1670
	19.	2-Arnino-5-ethyl-3-nitro-benzoic acid	$C_9H_{10}N_2O_4$	210	2070
	20.	Eugenyl acetate	$C_{12}H_{14}O_3$	206	1552
	21.	3-Allyl-6-methoxyphenyl acetate	$C_{12}H_{14}O_3$	206	1552
	22.	Eugenol (2-methoxy-4-(2-propenyl)	$C_{10}H_{12}O_2$	164	1392
	23.	Caryophyllene oxide	$C_{15}H_{24}O$	220	1507

(Hemalatha et al., 2016). Eugenyl acetate was present in the CID sample. The samples also included beta-caryophyllene, a sesquiterpene that is frequently present in clove oil and is well-known for its analgesic and anti-inflammatory effects (Amelia et al., 2017). The oil also contains minor amounts of sesquiterpene hydrocarbons, oxygenated monoterpenes, and other phenolic chemicals (Oliveira et al., 2016). In this study, phenolic chemicals are only identified in CID. For certain phenolic compounds, GC-MS may not always provide adequate sensitivity and specificity, particularly when they are present in complex mixes or in low levels (Proestos et al., 2006).

Concentration (ppm) of TFC, TPC and TAC in three brands cloves

CEO is acknowledged for its potent antioxidant capabilities. The principal constituent responsible for this activity is eugenol, which comprises approximately 76.8% in clove essential oil (Jirovetz et al., 2006). The antioxidant activity of the clove powder sample was measured as 70.66 ppm, 70.14 ppm, and 69.74 ppm for the CIS, CID, and CSI clove essential oils, respectively, as presented in Table 4. The contents are consistent with prior research (Alfikri et al., 2020). Besides eugenol, other constituents including β-caryophyllene and α-humulene further enhance the oil's antioxidant properties. The oil's capacity to mitigate oxidation reactions and diminish free radicals renders it a significant potential for pharmaceutical formulations targeting oxidative stress and associated disorders (Kiki, 2023). The TFC of the clove essential oil was measured as 244.36 ppm, 161.13 ppm, and 321.95 ppm for the CIS, CID, and CSI varieties, respectively. Although comprehensive quantitative data on the total flavonoid concentration of clove essential oil is limited in the existing literature, clove is recognized as a significant source of bioactive constituents, encompassing flavonoids and phenolic compounds (Pandey et al., 2023). The TPC of the clove essential oils was measured as 159.44 ppm, 142.82 ppm, and 135.11 ppm for the CIS, CID, and CSI varieties, respectively. A robust association frequently exists between the total phenolic content (TPC) of a plant extract and its antioxidant activity. Elevated concentrations of phenolic compounds generally signify enhanced antioxidant (Zargoosh et al., 2019). The average TAC values were determined to be 16.07 mg/100 g, 18.156 mg/100 g, and 17.94 mg/100 g for the CIS, CID, and CSI clove essential oils, respectively. The CEO is not recognized for its anthocyanin content. The oil is predominantly derived from the flower buds and leaves of the clove plant, resulting in a minimal anthocyanin concentration.

Zone of inhibition (mm) showing the antibacterial activity of CEO

The results obtained for *E. coli*, *Staphylococcus aureus* and *Pseudomonus aeruginosa* observed using the Kirby-Bauer disk diffusion method are illustrated in Table 5. In all experiments, the negative controls showed no zone of inhibition.

Table 4. Concentration (ppm) of TFC, TPC, TAC, and AOA in cloves.

Sample ID	TFC	TPC	TAC	AOA
CIS	244.36±0.01^{b}	159.44±0.01^{a}	16.071±0.01^{c}	70.66 ±0.01^{a}
CID	161.13±0.01^{c}	142.82±0.01^{b}	18.156±0.01^{a}	70.14±0.01^{b}
CSI	321.95±0.01^{a}	135.11±0.01^{c}	17.942±0.01^{b}	69.74±0.01^{c}

Mean ± S.D. for three replicates (n=3) and different superseripts indicate differences among orgin of clove. TFC= total flavonoid content; TPC= total phenolic content; TAC= total antioxidant capacity; AOA= antioxidant activity.

Table 5: Antibacterial activity of clove essential oil extract (100 µg/ml)

Country of origin	*Escherichia coli*		*Staphylococcus aureus*		*Pseudomonus aeruginosa*	
	Essential Oil (mL)	Positive Control (mm)	Essential Oil (mL)	Positive Control (mm)	Essential Oil (mL)	Positive Control (mm)
CIS	25	Nz	26	30	26	36
CID	24	Nz	25	28	25	27
CSI	27	Nz	24	29	24	33

CIS = Indonesian origin; CID = Indian origin; CSI = Sri Lanka origin; Nz = No zone of inhibition; Positive control: Ciprofloxacin

Eugenol is a phenolic substance. Phenols possess antibacterial characteristics (Nuñez and Aquino, 2012). Findings indicate that *S. aromaticum* contains bioactive compounds, underscoring its significance as a therapeutic plant. The results indicate that *S. aromaticum* extracts impede bacterial proliferation. The most substantial inhibitory zone measured 27 mm against *E. coli* by CSI. The clove oil extract showed antibacterial activity against all tested Gram-positive and Gram-negative pathogens. Previous studies demonstrate a comparable pattern

obstructing the proliferation of several pathogens,

such as *Staph. aureus, E. coli*, and *P. aeruginosa* (Kovács et al., 2016). No significant difference was seen among CIS, CID, and CSI regarding their bactericidal efficacy. All of them possessed the capacity to limit the proliferation of subsequent infections. Nonetheless, the clove oil extract exhibited greater efficacy against the *Staph. aureus* isolate at doses of 100 µg/ml. Ciprofloxacin exhibited no effect against *E. coli*, whereas the clove oil extract showed significant efficacy against *E. coli*. The antibacterial efficacy is affected by variables including oil content, temperature, and the presence of organic materials (Nuñez and Aquino, 2012).

Conclusion

Lower acid and peroxide value in CIS, CID, and CSI indicate enhanced stability and extended shelf-life. The viscosity (η) of the CEOs exhibited a linear correlation with temperature. The GC-MS analysis of the oil extract identified eugenol, caryophyllene, and eugenol acetate as the principal constituents. The antibacterial properties of clove oil extract can be attributed to the discovered components. Eugenol and caryophyllene are well-known for their antibacterial effects. Consequently, impeding the advancement and proliferation of all examined pathogens and indicator organisms, particularly *E. coli, Staph. aureus*, and *P. aeruginosa*. The most significant inhibitory impact of clove oil was observed against *E. coli* by CSI. However, some asymmetry was also observed due to environmental factors such as temperature, photosynthetic rate, and heat, which influence the carbohydrate and protein levels in cloves. All three varieties of cloves exhibited no significant differences among them. The study suggested that clove and clove oils may serve as antibacterial and antiseptic agents. Subsequent research may involve developing goods including clove essential oil to enhance their longevity.

Acknowledgment

Department of Applied Chemistry and Chemical Technology, CVASU for technical assistance and material support.

Authors contribution

S.H. and I.H. performed the experiments and collected data. S.A. and A.C. analyzed the results and prepared the manuscript draft. M.T.A. and S.C. contributed to data interpretation and literature review. M.A. supervised the study, provided critical insights, and approved the final manuscript.

Conflict of interest

The authors declare no conflicts of interest.

References

Akther S, Jothi JS, Badsha MR., Rahman MM, Das GB and Alim MA. Drying methods effect on bioactive compounds, phenolic profile, and antioxidant capacity of mango powder. *J. King Saud Univ. Sci.*, 2023; 35(1): 102370.

Alanazi AK, Alqasmi MH, Alrouji M, Kuriri FA, Almuhanna Y, Joseph B and Asad M. Antibacterial activity of syzygium aromaticum (Clove) bud oil and its interaction with imipenem in controlling wound infections in rats caused by methicillin-resistant staphylococcus aureus. *Molecules*, 2022; 27(23): 8551.

Alfikri FN, Pujiarti R, Wibisono MG and Hardiyanto EB. Yield, quality, and antioxidant activity of clove (*Syzygium aromaticum* L.) bud oil at the different phenological stages in young and mature trees. *Scientifica*, 2020; 2020: 9701701.

Alma MH, Ertaş M, Nitz S and Kollmannsberger H. Chemical composition and content of essential oil from the bud of cultivated Turkish clove (*Syzygium aromaticum* L.). *Bio Resou.*, 2007; 2(2): 265-269.

Al-Owaisi M, Al-Hadiwi N and Khan SA. GC-MS analysis, determination of total phenolics, flavonoid content and free radical scavenging activities of various crude extracts of *Moringa peregrina* (Forssk.) Fiori leaves. *Asian Pac. J. Trop. Biomed.* 2014; 4(12): 964-970.

Amelia B, Saepudin E, Cahyana AH, Rahayu DU, Sulistyoningrum AS and Haib J. GC-MS analysis of clove (Syzygium aromaticum) bud essential oil from Java and Manado: 2nd International symposium on current progress in mathematics and sciences 2016, ISCPMS 2016. *International Symposium on Current Progress in Mathematics and Sciences*. 2017; 2016: ISCPMS 2016.

Aziz S, Naher S, Abukawsar M and Roy SKB. Comparative studies on physicochemical properties and GC-MS analysis of essential oil of the two varieties of the black pepper (Piper nigrum Linn.). *Int. J. Pharm. Phytopharm. Res.* 2012; 2(2): 67-70.

Bhuiyan MNI, Begum J, Ch N, NR I and Akter F. Constituents of the essential oil from leaves and buds of clove (Syzigium caryophyllatum (L.) Alston). *Afr. J. Plant Sci.* 2010; 4(11): 451-454.

Chang CC, Yang MH, Wen HM and Chern JC. Estimation of total flavonoid content in propolis by two complementary colometric methods. *J. Food Drug Anal.* 2002; 10(3): 178-182.

Drew WL, Barry ALO, Toole R and Sherris JC. Reliability of the kirby-bauer disc diffusion method for detecting methicillin-resistant strains of staphylococcus aureus. *Appl. Microbiol.* 1972; 24(2): 240-247.

Gandova V, Lazarov A, Petkova N and Stoyanova A. Physicochemical parameters and antioxidant activity of summer savory essential oil (*Satureja hortensis* L.)-Consensus. *BIO Web of Conferences*, 2024; 102: 01017.

Gharby S, Harhar H, Bouzoubaa Z, Asdadi AEl, Yadini A and Charrouf Z. Chemical characterization and oxidative stability of seeds and oil of sesame grown in Morocco. *J. Saudi Soc. Agric. Sci.* 2017; 16(2): 105-111.

Haro-González JN, Castillo-Herrera GA, Martínez-Velázquez M and Espinosa-Andrews H. Clove essential oil (*Syzygium aromaticum* L. Myrtaceae): extraction, chemical composition, food applications, and essential bioactivity for human health. *Molecules*, 2021; 26(21): 6387.

Hemalatha R, Nivetha P, Mohanapriya C, Sharmila G, Muthukumaran C and Gopinath M. Phytochemical composition, GC-MS analysis, in vitro antioxidant and antibacterial potential of clove flower bud (*Eugenia caryophyllus*) methanolic extract. *J. Food Sci. Technol.*, 2016; 53(2): 1189-1198.

Jayarathne SK, Liyanage T and Kumari DWMMM. Physical, chemical and morphological characterization of Sri Lankan origin dried clove (*Syzygium aromuticum* L.) BUDS. 2020.

Jirovetz L, Buchbauer G, Stoilova I, Stoyanova A, Krastanov A and Schmidt E. Chemical composition and antioxidant properties of clove

leaf essential oil. *J. Agri. Food Chem.,* 2006; 54(17): 6303-6307.

Kiki MJ. In Vitro antiviral potential, antioxidant, and chemical composition of clove (*Syzygium aromaticum*) essential oil. *Molecules*, 2023; 28(6): 2421.

Kovács JK, Felső P, Makszin L, Pápai Z, Horváth G, Ábrahám H, Palkovics T, Böszörményi A, Emődy L and Schneider G. Antimicrobial and virulence-modulating effects of clove essential oil on the foodborne pathogen *campylobacter jejuni*. *Appl. Environ. Microbiol.* 2016; 82(20): 6158-6166.

Kyriakidis NB and Katsiloulis T. Calculation of iodine value from measurements of fatty acid methyl esters of some oils: Comparison with the relevant American Oil Chemists Society method. *J. Am. Oil Chem. Soc.*, 2000; 77(12): 1235-1238.

Liñán-Atero R, Aghababaei F, García SR, Hasiri Z, Ziogkas D, Moreno A and Hadidi M. Clove essential oil: chemical profile, biological activities, encapsulation strategies, and food applications - consensus. *Antioxidants*, 2024; 13(4): 488.

Liu H, Schmitz JC, Wei J, Cao S, Beumer JH, Strychor S, Cheng L, Liu M, Wang C, Wu N, Zhao X, Zhang Y, Liao J, Chu E and Lin X. Clove extract inhibits tumor growth and promotes cell cycle arrest and apoptosis. *Oncol. Res.* 2014; 21(5): 247-259.

Mohan R, Jose S, Mulakkal J, Karpinsky-Semper D, Swick AG and Krishnakumar IM. Water-soluble polyphenol-rich clove extract lowers pre- and post-prandial blood glucose levels in healthy and prediabetic volunteers: An open label pilot study. *BMC Complement. Altern. Med.* 2019; 19(1): 99.

Nuñez L and Aquino MD. Microbicide activity of clove essential oil (*Eugenia caryophyllata*). *Brazilian J. Micro.*, 2012; 43(4): 1255-1260.

Nzeako BC, Al-Kharousi ZSN and Al-Mahrooqui Z. Antimicrobial activities of clove and thyme extracts. *Sultan Qaboos Univ. Med. J.* 2006; 6(1): 33-39.

Oliveira M, Costa WA, Pereira DS, Botelho JRS, Menezes TO, Andrade E, Silva SEM, Filho AP da SS and Junior RC. Chemical composition and phytotoxic activity of clove (*Syzygium aromaticum*) essential oil obtained with supercritical CO2—Consensus. *J. Supercrit. Fluids*, 2016; 118: 185-193.

Paez V, Barrett WB, Deng X, Diaz-Amigo C, Fiedler K, Fuerer C, Hostetler GL, Johnson P, Joseph G, Konings EJM, Lacorn M, Lawry J, Liu H, Marceau E, Mastovska K, Monteroso L, Pan SJ, Parker C, Phillips MM and Coates SG. AOAC SMPR® 2016.002. *J. AOAC Inter.*, 2016; 99(4): 1122-1124.

Pandey VK, Srivastava S, Ashish Dash KK, Singh R, Dar AH, Singh T, Farooqui A, Shaikh AM and Kovacs B. Bioactive properties of clove (Syzygium aromaticum) essential oil nanoemulsion: A comprehensive review. *Heliyon*, 2023; 10(1): e22437.

MHPRA (Medicines and Healthcare products Regulatory Agency). Pharmacopoeia Commission: New arrangements for human medicines in line with the Windsor Framework, 2011.

Porter NG and Lammerink JP. Effect of temperature on the relative densities of essential oils and water—consensus. *J. Essent. Oil Res*. 1994; 6: 269-277.

Proestos C, Sereli D and Komaitis M. Determination of phenolic compounds in aromatic plants by RP-HPLC and GC-MS. *Food Chem.* 2006; 95(1): 44-52.

Shavisi N, Akhondzadeh Basti A, Khanjari A, Misaghi A, Shahbazi Y, Hajjar Bargh A and Vanaki E. In Vitro antibacterial activity of polylactic acid film incorporated with ethanolic propolis extract and ziziphora clinopodioides essential oil. *J. Food Qual. Hazards Control*, 2017: 4: 3-8.

Srivastava K and Malhotra N. Acetyl eugenol, a component of oil of cloves (Syzygium aromaticum L.) inhibits aggregation and alters arachidonic acid metabolism in human blood platelets. *Prostaglandins Leukot. Essent. Fat. Acids*. 1991; 42(1): 73-81.

Sulieman EA, El Boshra I and El Khalifa EA. Nutritive value of clove (*Syzygium aromaticum*) and detection of antimicrobial effect of its bud oil. *Res. J. Microbiol.* 2007; 2(3): 266-271.

Takeda J, Silva S, Kawet L and Lanka S. Spices in Sri Lanka, India and Bangladesh with special reference to the usages and consumptions. *Bull. Fac. Agr. Saga Univ.* 2008; 93: 1-25.

Tomaino A, Cimino F, Zimbalatti V, Venuti V, Sulfaro V, Pasquale A and Saija A. Influence of heating on antioxidant activity and the chemical composition of some spice essential oils—Consensus. *Food Chem.* 2005; 89: 549-554.

Zahir E, Saeed R, Hameed MA and Yousuf A. Study of physicochemical properties of edible oil and evaluation of frying oil quality by Fourier Transform-Infrared (FT-IR) Spectroscopy. *Arabian J. Chem.*, 2017; 10: S3870-S3876.

Zargoosh Z, Ghavam M, Bacchetta G and Tavili A. Effects of ecological factors on the antioxidant potential and total phenol content of Scrophularia striata Boiss. *Sci. Rep.* 2019; 9(1): 16021.