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Chemical composition and biological activities of acetone extract from the seed peel of *Madhuca elliptica*

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ABSTRACT

This study explores the chemical composition and biological activities of the acetone extract derived from the seed peel of *Madhuca elliptica* (Sapotaceae family). Located in the Binh Chau - Phuoc Buu Nature Reserve of Vietnam, the seeds of *M. elliptica* were subjected to a methodical extraction process using acetone, followed by chemical analysis through Gas Chromatography-Mass Spectrometry (GC-MS). The analysis identified 35 distinct compounds, with a notable presence of cis-13 octadecenoic acid, n-hexadecanoic acid, octyl, diethyl phthalate, and octadecanoic acid. The biological efficacy of the extract was evaluated through two main bioassays: antioxidant and antibacterial activities. The antioxidant capacity was determined using the ABTS radical scavenging method, revealing a significant correlation between the concentration of the extract and its ability to neutralize free radicals, with an IC_{50} value of 6.259 ppm. Additionally, the antibacterial activity was assessed using the Kirby-Bauer disk diffusion method against a panel of both Gram-positive and Gram-negative bacteria. Through this research, *M. elliptica* exhibits significant potential due to its bioactive compounds, suggesting its applicability in pharmaceutical and nutraceutical fields. Besides, this study adds valuable insights into the utilizable aspects of *M. elliptica*, encouraging further research into its diverse applications.

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INTRODUCTION

The family Sapotaceae comprises 1250 species of flowering trees and/or shrubs spanning 53 genera and five tribes, predominantly concentrated in the tropical and subtropical regions of Asia and South America, with notable diversity (Swenson & Anderberg, 2005; Baky *et al.*, 2022). Sapotaceae yield a range of products including plant latex (e.g., from Manilkara zapota), valuable timber (e.g., *Madhuca pasquieri*), and diverse fruit-bearing trees (Pennington, 2004; Kubitzki, 2013; Baky *et al.*, 2022). In Vietnam, approximately 50 species from this family have been identified. At present, based on phylogenetic analyses and morphological assessments, the Sapotaceae family is taxonomically divided into three monophyletic subfamilies: *Sarcospermatoideae*, *Sapotoideae*, and *Chrysophylloideae*. Among them, *Pouteria* and *Chrysophyllum* represent the two largest genera within the Sapotaceae (Alves-Araújo *et al.*, 2020).

The genus *Madhuca*, first described in 1791, originates from South Asia, East Asia, Southeast Asia, and Papuasia, encompassing species such as *Madhuca alpina*, *Madhuca butyrospermoides*, *Madhuca pasquieri* (listed in the Vietnam Red Book in 1996 due to intense illegal exploitation) (WCMC, 1998), *Madhuca longifolia*, *Madhuca indica*, and others (GBIF, 2024). Currently, there are approximately 116 recognized species within the *Madhuca* genus (GBIF, 2024). There have been numerous reports indicating the presence of valuable compounds, particularly low molecular weight substances including saponins, carbohydrates, triterpenoids, steroids, flavonoids, and glycosides isolated from various *Madhuca* species. Carotenoids, predominantly found in the seeds of *Madhuca* species, have applications in medicine for treating

skin diseases, arthritis, headaches, and as laxatives. Furthermore, the seeds of *Madhuca* species have been proven to contain high lipid content, which can be utilized in the production of soap, lubricants, and biodiesel (Phuong *et al.*, 2019).

Madhuca elliptica, also known as *Dasyaulus elliptica* Pierre ex Dubard or *Payena elliptica* (Pierre ex Dubard) Lecomte, is commonly referred to as "viết", "sến bầu dục", or "sến núi đinh" in Vietnamese (BVN, 2024). Morphological characteristics include narrowly elliptic leaves with no hair, 9-13 pairs of secondary veins, and long petioles measuring 10-15 mm. The flowers form clusters in the leaf axils, with fine hairs present. Each flower comprises 4 hairy sepals, measuring 4 mm in height, and 8 lobes in the corolla tube, without hair. The elongated ovoid fruit measures 3-3.5 cm in length, containing a single seed measuring 2 cm (Pham, 1991).

Although there is limited research specifically on *M. elliptica*, studies on the genus Madhuca have yielded promising results. For instance, *M. longifolia*, another species within the genus, has been utilized in synthesizing green color nanocomposite pigments using plant extracts, demonstrating effective wastewater treatment and antibacterial properties. Nano CuO particles synthesized from *M. longifolia* extract exhibit excellent antibacterial activity against bacterial strains such as *E. coli*, *S. aureus*, and *B. subtilis*, highlighting the antibacterial potential of this species (Das *et al.*, 2018). Roy *et al.* (2010) demonstrated the considerable antioxidant properties of the ethanolic extract derived from the bark of *M. longifolia*. Furthermore, research by Chaudhary *et al.* (2012) explored the antioxidant and free radical scavenging activity of methanol extracts from *M. indica* bark, suggesting its potential medical use as an effective antioxidant. Results indicate that the high content of phenolic compounds in the extract may contribute to its antioxidant activity, as phenolic compounds are known for their direct antioxidant properties due to the presence of hydroxyl groups, which can act as hydrogen donors (Chaudhary *et al.*, 2012).

This study aims to investigate the chemical composition and biological activities, including antioxidant and antibacterial properties, of the acetone extract from the seed peel of *M. elliptica*.

MATERIALS AND METHODS

Plant Material

Seeds samples of *Madhuca elliptica* were collected from the Binh Chau - Phuoc Buu Nature Reserve, Bung Rieng commune, Xuyen Moc district, Ba Ria-Vung Tau province, Vietnam. The scientific name of the studied species was determined by Van Son Le and the voucher specimen, VS Le 1012A, was deposited at the herbarium of this Nature Reserve.

Bacterial Strain

Two strains of Gram-positive bacteria (*Bacillus cereus* ATCC 11774 and *Staphylococcus aureus* ATCC 25923) and four strains of Gram-negative bacteria (*Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Salmonella enteritidis* ATCC 13976, and *Salmonella typhimurium* ATCC 13311) were used to determine the antibacterial activity of the studied extract.

Extraction Method

Seed peels of *M. elliptica* were dried at 50 °C until a constant weight was achieved and it was then ground into powder. One hundred grams of the powdered seed peel was weighed and immersed in 500 mL of acetone at room temperature for 72 hours. The mixture was filtered through Whatman filter paper, and the extract was collected. An additional 300 mL of acetone was added to the residue, and followed by a second filtration. The extract was concentrated under vacuum pressure at 45 °C to collect the brown extract.

Qualitative Analysis of Natural Compounds

The presence of some natural compounds in the acetone extract from the seed peel of *M. elliptica* was determined as described in Table 1.

GC-MS based Chemical Profiling of the Acetone Extract

The chemical analysis of the acetone extract from the species under study was conducted using a TRACETM 1310 Gas Chromatograph, manufactured by Thermo Fisher Scientific Inc., based in Waltham, MA, USA. This equipment included an ISQ 7000 single quadrupole mass spectrometer and a DB-5MS column measuring 30 m x 0.25 mm x 0.25 µm. The samples were introduced into the GC system, which was maintained at 250 °C and a split ratio of 30:1, with a flow rate of 36 mL/min and a splitless duration of 1 minute. Helium served as the carrier gas, flowing at 1.2 mL/min. The temperature of the transfer line was also maintained at 250 °C. Initially, the oven temperature was stabilized at 80 °C for 5 minutes before escalating to 280 °C at a rate of 20 °C/min, where it remained for 10 minutes. Electron impact ionization was conducted at 70 eV, and the filament source temperature was 250 °C. The mass spectrometer's acquisition scanned a mass range from 29 to 650 m/z at a rate of 2 scans per second. The identification of chemical components within the acetone extracts from *M. elliptica* was achieved by matching their mass spectra with the NIST 2017 library (Van *et al.*, 2023).

ABTS Free Radical Scavenging Assay

The procedure described here is a revised version of the one outlined by Re *et al.* (1999). Initially, a 7 mM ABTS (2,2′-Azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) solution was combined with a 2.45 mM potassium persulfate solution and left to stand in the dark for 16 hours to produce the ABTS radical cation solution. This solution was then diluted with 50% methanol to achieve an initial optical density of roughly 0.70 ± 0.02 at 745 nm, prior to its application in the experimental assays. For the assay, 3 mL of the prepared ABTS solution was mixed with 300 µL of the sample under investigation in a microcuvette to assess its antioxidant capacity. The absorbance was measured

Table 1: Qualitative analysis methods of natural compounds

Compounds	Methods	Positive reaction results	References
Phenolic and tannin	4 mL seed peel extract sample + few drops of 0.1% $FeCl_3$	Brownish green or blue-black coloration appears	Sankhalkar & Vernekar, 2016; Gul *et al.*, 2017; Dahanayake *et al.*, 2019
Flavonoid	2 mL seed peel extract sample + 3 mL of dilute ammonia + 1 mL conc. H_2SO_4	Yellow coloration appears	Dahanayake *et al.*, 2019
Quinone	2 mL seed peel extract sample + 5 mL HCl	Yellow colored precipitate appears	Muniyandi & Lakshman, 2018
Coumarin	2 mL seed peel extract sample + 3 mL NaOH (10%)	Solution turns to yellow colour	Muniyandi & Lakshman, 2018
Alkaloid	2 mL seed peel extract sample + two drops of Wagner reagent	Reddish coloration appears	Dahanayake *et al.*, 2019
Terpenoid	3 mL seed peel extract sample + 1 mL $CHCl_3$ + 1 mL conc. H_2SO_4 (Salkowski test)	Red-brown coloration appears	Gul *et al.*, 2017; Dahanayake *et al.*, 2019
Saponin	5 mL seed peel extract sample + 2.5 mL distilled water + three drops of saturated oil + boiled and vigorously shaken	Emulsion formed	Gul *et al.*, 2017; Dahanayake *et al.*, 2019

after six minutes (Re *et al.*, 1999; Noufal *et al.*, 2023). The percentage inhibition was calculated using the Equation 1:

$$\% \text{ of cell inhibition} = [(\text{ODcontrol} - \text{ODsample})/\text{ODcontrol}] \times 100 \quad (1)$$

From the percentage neutralization (%) obtained by the ABTS, a linear correlation equation y=ax+b was constructed. Using this equation, the IC_{50} value is determined by substituting y=50, thereby finding the corresponding x value, which represents the concentration at which 50% of the ABTS free radicals are neutralized.

Antibacterial Activity

The antibacterial activity of the acetone extract from the seed peel of *Madhuca elliptica* was determined using the Kirby-Bauer disk diffusion method. Bacterial strains were cultured in Mueller-Hinton Broth (MHB) until reaching a turbidity of 0.5 McFarland standard. Subsequently, a 0.1 mL aliquot of the bacterial suspension was inoculated onto Mueller-Hinton Agar (MHA) plates and evenly spread. Paper discs were then positioned on the agar surface, onto which 10 µL of the sample solution was applied. Gentamycin antibiotic discs (Nam Khoa, Vietnam) served as positive controls. The Petri dishes were refrigerated at 4 °C for 2 hours to allow the sample solution to permeate into the agar. Following this, the cultures were incubated at 37 °C for 16-18 hours to assess the extract's resistance against the bacterial strains. The diameter of the inhibition zone was measured after 16-18 hours. The antibacterial activity of the sample solutions against the bacterial strains was determined based on the diameter of the inhibition zones, with the diameter of the paper disc (6 mm) subtracted to obtain the zone of bacterial inhibition for the research sample (Hindler *et al.*, 2006).

RESULTS AND DISCUSSION

Qualitative Analysis of Natural Compounds of Acetone Extract from the Seed Peel of *Madhuca elliptica*

Figure 1 depicted the powder and acetone extract obtained from the seed peel of *M. elliptica*, with the acetone extract being characterized by a brownish color.

The results of qualitative analysis (Table 2) suggested that phenolics, tannins, flavonoids, alkaloids, and terpenoids were present in the seed peel of *M. elliptica*.

These compounds have been extensively studied for their confirmed biological roles, which include antibacterial and antioxidant activities (Borges *et al.*, 2013; Russo *et al.*, 2013; Kaurinovic & Vastag, 2019; Muniyandi *et al.*, 2019).

According to the research conducted by Singh *et al.* (2018), during the quantification of certain compounds from *M. longifolia* species harvested from Uttarakhand, India, acetone extraction from the oil cakes of *M. longifolia* revealed the presence of alkaloids (using Dragendroff's test and Mayer's test), phenolic compounds, and tannins (using Ammonia test and Ferric chloride test), saponins (using Foam test and Lead Acetate test), and terpenoids (utilizing Salkowski test) (Singh *et al.*, 2018). Furthermore, in the qualitative analysis of natural compounds from the ethanol extract of *M. longifolia* leaves collected from Uttar Pradesh, India, Khare *et al.* (2017) demonstrated the positive reactions of various compounds such as steroids, saponins, flavonoids, tannins, and triterpenoids. Additionally, acetone extraction from the fresh inner bark of *M. indica* collected from Andhra Pradesh, India, was found to contain alkaloids and saponins (Gujjeti & Mamidala, 2013).

The Chemical Composition of the Acetone Extract from the Seed Peel of *Madhuca elliptica*

The Gas Chromatography-Mass Spectrometry (GC-MS) analysis results of acetone extract from the seed peel of *M. elliptica* collected in Binh Chau - Phuoc Buu revealed the identification of 35 compounds, as presented in Table 3.

From the chromatogram (Figure 2), the concentration of compounds in the sample can be observed with their corresponding retention times. Higher peaks indicate higher concentrations of compounds in the sample. Some compounds exhibit high concentrations in the acetone extract from the seed peel of *M. elliptica*, such as cis-13 octadecenoic acid with a retention time (RT) of 14.86; n-hexadecanoic acid (RT=13.96); octyl (RT=16.98); diethyl phthalate (RT=11.85); octadecanoic acid (RT=14.94).

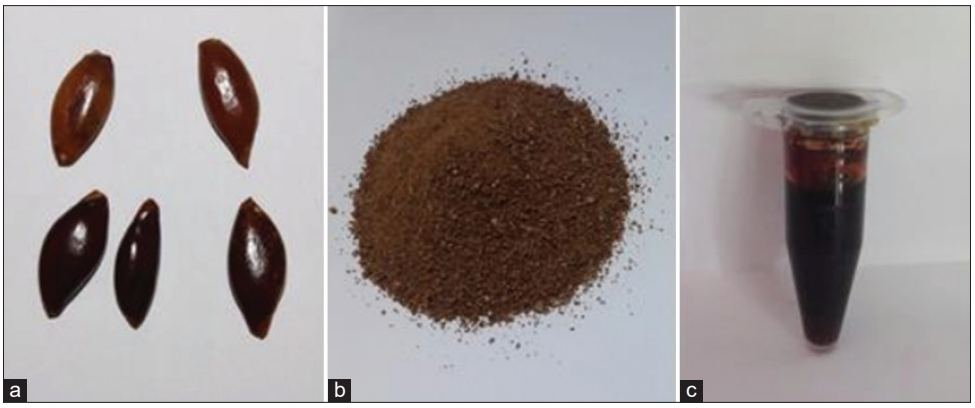

Figure 1: Acetone extract from the seed peel of *M. elliptica* (a) Seed, b) Powder from seed peel and c) Acetone extract from seed peel)

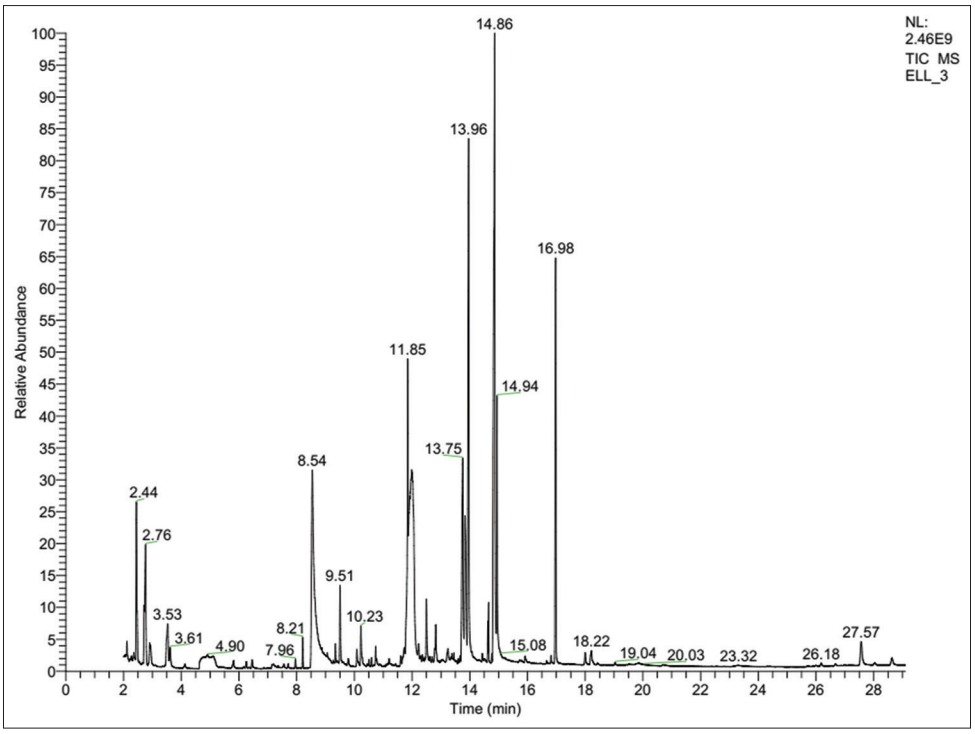

Figure 2: Gas chromatogram of acetone extract from *M. elliptica*

Table 2: Qualitative analysis of natural compounds of acetone extract from the seed peel of *Madhuca elliptica*

No.	Compounds	Positive/negative
1	Phenolic and tannin	+
2	Flavonoid	+
3	Quinone	-
4	Coumarin	-
5	Alkaloid	+
6	Terpenoid	+
7	Saponin	-

Studies have explored the chemical constituents derived from different solvent extracts of various Madhuca species. For example, the chemical composition of leaf extracts from *M. longifolia* (Koenig) Linn, collected from Tamil Nadu, India, was analyzed using gas chromatography-mass spectrometry (GC-MS). This analysis identified approximately 20 components. Predominantly, heterocyclic compounds such as furfural, imidazoles, furan carboxylic acid, piperidine, pyrazole, chromones, and pyrrolidone were detected. Among these, furan and pyrrolidine compounds exhibited the highest percentages in the peak areas, specifically 22.5% for 2-Furancarboxaldehyde, 5-(hydroxymethyl)- and 22.27% for 1-Methyl-2-pyrrolidone-4-carboxamide. Additional notable components included α-D-Mannofuranoside, methyl (19.7%); 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl- (12.1%); and others (Annalakshmi *et al.*, 2013).

According to the study by Souravi *et al.* (2015), the methanol extracts from the leaves, roots, bark, fruits, and seeds of *M. insignis*, which were identified and collected in Karnataka, India, underwent analysis through gas chromatography-mass spectrometry (GC-MS). The GC-MS profiling revealed a total of 31 known

Table 3: The chemical composition of acetone extract from the seed peel of *M. elliptica*

Retention time (min)	Compounds	Molecular formula
2.44	Butyl acetate	$C_6H_{12}O_2$
2.76	Acetonyldimethylcarbinol	$C_6H_{12}O_2$
3.53	Butyl oxitol	$C_6H_{14}O_2$
3.61	Dimethyl sulfone	$C_2H_6O_2S$
5.11	Glycerin	$C_3H_8O_3$
5.82	Monomethyl malonate	$C_4H_6O_4$
6.26	Benzyl alcohol	C_7H_8O
7.70	Isophorone	$C_9H_{14}O$
7.96	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl	$C_6H_8O_4$
8.21	Acetic acid, phenylmethyl ester	$C_9H_{10}O_2$
8.54	Catechol	$C_6H_6O_2$
9.34	Hydroquinone	$C_6H_6O_2$
9.51	1,2-Benzenediol, 4- methyl	$C_7H_8O_2$
10.08	Phenol, 2,6-dimethoxy	$C_8H_{10}O_3$
10.23	Pyrogallol	$C_6H_6O_3$
10.50	Vanillin	$C_8H_8O_3$
10.59	Diphenyl ether	$C_{12}H_{10}O$
10.74	4-Methoxybenzene-1,2-diol	$C_7H_8O_3$
11.85	Diethyl phthalate	$C_{12}H_{14}O_4$
12.00	Phloroglucinol	$C_6H_6O_3$
12.50	Benzo[b] tetrahydrofuran-3-one, 5,6-dihydroxy	$C_8H_6O_4$
12.82	Tetradecanoic acid	$C_{14}H_{28}O_2$
13.72	1,3-Diethoxybenzene	$C_{10}H_{14}O_2$
13.85	2H-1-Benzopyran-2-one, 3,5,7-trihydroxy	$C_9H_6O_5$
13.96	n-Hexadecanoic acid	$C_{16}H_{32}O_2$
14.62	Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$
14.65	trans-13-Octadecenoic acid, methyl ester	$C_{19}H_{36}O_2$
14.82	9(E),11(E)-Conjugated linoleic acid	$C_{18}H_{32}O_2$
14.86	cis-13-Octadecenoic acid	$C_{18}H_{34}O_2$
14.94	Octadecanoic acid	$C_{18}H_{36}O_2$
16.98	Octoil	$C_{24}H_{38}O_4$
18.00	(E)-3,3'-Dimethoxy-4,4' dihydroxystilbene	$C_{16}H_{16}O_4$
18.22	9-Octadecenoic acid (Z)-, 2-hydroxy-1-hydroxymethyl ethyl ester	$C_{21}H_{40}O_4$
27.57	Stigmasterol	$C_{29}H_{48}O$
28.62	β-Sitosterol	$C_{29}H_{50}O$

compounds. The diverse classes of these identified compounds ranged from fatty acid derivatives, steroids, and terpenoids to heterocyclic compounds, carotenoids, and phenolic compounds. Notable compounds included Germanicol, found in the root, fruit, and seed; 7-Oxocholest-5-en-3-yl benzoate, derived from the leaf; alpha Tocopherol/Vitamin E, present in the leaf, bark, root, and fruit; 1-hexacosene, from the leaf; and 2,3-Dihydroxypropyl elaidate, also from the leaf, among others (Souravi *et al.*, 2015).

Antioxidant Activity of Acetone Extract from the Seed Peel of *M. elliptica*

The percentage neutralization (%) obtained by the ABTS of the acetone extract from *M. elliptica* was calculated and is presented in Table 4. Using the equation y=7.1187x+5.447, the IC_{50} value was determined to be 6.259±0.24.

From the results depicted in Figure 3, the antioxidant activity of the acetone extract from the seed shell of *M. elliptica* is directly proportional to the concentration of the extract. As the concentration of acetone extract from *M. elliptica* increases

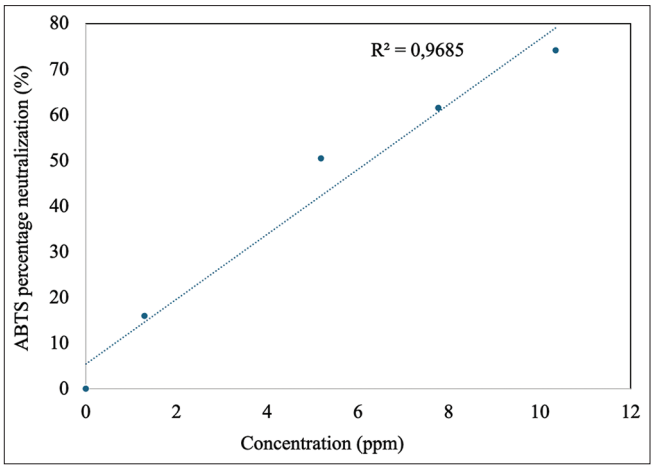

Figure 3: The percentage neutralization (%) obtained by the ABTS of acetone extract from *M. elliptica*

from 0 to 10.35 (ppm), the free radical scavenging efficiency also increases from 0% to 78.49%. The IC_{50} value of the acetone extract sample from *M. elliptica* was determined to be 6.26. The observed antioxidative effect of the sample is likely attributed to the presence of phenolic and flavonoid compounds in the acetone extract. As indicated by the identification of certain natural compounds, both acetone extract samples contain flavonoid and phenolic compound groups, which are known for their potent antioxidative properties. Phenolic compounds, including flavonoids, are renowned for their exceptional antioxidative capabilities in plants (Madhavi *et al.*, 1995). According to many research, ABTS degradation partly results from the reaction with polyphenols such as flavonoids. Previous reports have also highlighted phenols and flavonoids as scavengers and free radical scavengers, possibly due to their molecular weight, the presence of aromatic rings, and the nature of their substituent OH groups (Sharma *et al.*, 2011; Zeb *et al.*, 2014).

The antioxidant activity of *M. longifolia* leaf extracts using ethyl alcohol, methanol, and aqueous solvents has been demonstrated. Among the solvents tested, the highest radical scavenging activity was observed in the following order: methanol > ethyl alcohol > aqueous (Bains *et al.*, 2020). Additionally, *M. longifolia* flowers and fruits (both ripe and unripe) collected from Uttar Pradesh, India, exhibited antioxidative properties. The ABTS radical scavenging capacities of *M. longifolia* flower extracts were ranked as follows: acetone > methanol > ethanol: methanol > water > ethanol fraction. The findings indicated that a 50% acetone solution exhibited the highest ABTS antioxidant activity for *M. longifolia* flowers, as well as unripe and ripe fruits (Singh *et al.*, 2017).

Antibacterial Activity of the Acetone Extract from the Seed Peel of *M. elliptica*

The antibacterial activity of the acetone extract from the seed peel of *M. elliptica* was investigated using the disc diffusion method against six bacterial strains. The antibacterial activity was demonstrated by the formation of clear zones of inhibition around the acetone extract sample in Table 5. From the

Table 4: Antioxidant Activity of acetone extract from the seed peel of *M. elliptica*

Concentration (ppm)	0	1.293	5.173	7.760	10.347
Average	0.372±0.02	0.312±0.02	0.183±0.03	0.144±0.05	0.097±0.02
ABTS percentage neutralization (%)	0	15.93±56.66	50.476±11.35	61.573±12.9	74.177±4.41

Table 5: The antimicrobial inhibition zone diameter of acetone extract from the seed peel of *M. elliptica*

Bacterial strains	Antimicrobial inhibition zone diameter (mm)
Bacillus cereus	10.3±2.1
Escherichia coli	-
Pseudomonas aeruginosa	-
Salmonella enteritidis	-
Salmonella typhimurium	11.7±2.1
Staphylococcus aureus	14.3±0.6

results in Table 5, the acetone extract of *M. elliptica* exhibited antibacterial activity against three bacterial strains: *S. aureus*, *S. typhimurium*, and *B. cereus*.

The acetone extract from the seed peel of *M. elliptica* demonstrated the highest antibacterial activity against the strain *S. aureus*, with an inhibition zone diameter of 14.3 mm. For the two strains *S. typhimurium* and *B. cereus*, the antibacterial efficacy of the acetone extract was weaker, with inhibition zone diameters of 11.7 mm and 10.3 mm, respectively. The remaining three strains, *E. coli*, *P. aeruginosa*, and *S. enteritidis*, showed little to no inhibition by the acetone extract.

Overall, both acetone extract samples exhibited the strongest antibacterial activity against the *S. aureus* strain. This may be attributed to the presence of identified natural compounds in *M. elliptica*, including flavonoids, alkaloids, terpenoids, phenolics, and tannins. Flavonoids impact *S. aureus* by inhibiting certain virulence factors of bacteria, including signal transduction receptors, enzymes, and toxins (Cushnie & Lamb, 2011), as well as RNA synthesis (Mori *et al.*, 1987). Terpenoids disrupt cell division and alter the morphology of *S. aureus* (Guimarães *et al.*, 2019). Additionally, alkaloid extracts possess selective antibacterial activity against Gram-positive bacteria such as *S. aureus* and *B. cereus*.

Furthermore, flavonoids bind to adhesion factors of Gram-negative bacteria and inhibit acetylcholine release - a component of the phospholipid layer, rendering them non-functional. Alkaloids penetrate bacterial cell walls, leading to bacterial structure disruption. Phenolic compounds, when in contact with Gram-negative bacteria, reduce metabolic activity, inhibit enzyme function, and damage bacterial cell membranes.

When assessing the antibacterial potential of *M. longifolia* species against two microbial strains, namely *S. pasteuri* (Gram-positive bacteria) and *P. aeruginosa* (Gram-negative bacteria), the extracts obtained from oil cake demonstrated antibacterial activity (Singh *et al.*, 2018). Guimarães *et al.* (2019) further substantiated the antibacterial efficacy of *M. longifolia* against strains of *S. aureus* and *E. coli* using leaf extracts prepared with ethyl alcohol, methanol, and aqueous solvents.

CONCLUSION

The research successfully analyzed the chemical composition and biological effects of the acetone extract derived from *M. elliptica* seed peel. Through GC-MS analysis, a diverse range of chemical compounds, mainly phenolic, flavonoid, alkaloid, and terpenoid substances, were identified. Notably, the extract demonstrated significant antioxidant properties, as demonstrated by the ABTS radical scavenging assay, indicating its ability to counteract free radicals effectively. Additionally, antibacterial tests revealed the extract's capability to inhibit bacterial growth, suggesting its potential in developing natural antibacterial remedies.

The findings from this study provide a foundation for further exploration into the pharmacological potential of *M. elliptica*. The identification of bioactive compounds opens new avenues for pharmaceutical applications, particularly in the development of antioxidant and antimicrobial agents. Furthermore, these results affirm the broader significance of the *Madhuca* genus in traditional and modern medicine, reinforcing the need for conservation of these species due to their medicinal value.

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