

PHOTOCHEMICAL, ANTIOXIDANT, AND ANTIMICROBIAL PROPERTIES OF BANANA PEEL CRUDE EXTRACT ON MANGO FRUIT SPOILING FUNGUS



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Abstract:	Phytochemicals derived from fruits and vegetables have garnered significant attention due to their diverse health benefits, including antioxidant, antimicrobial, and anti-inflammatory properties. Banana (Musa spp.) peel, an underutilized agricultural byproduct, contains bioactive compounds offering potential for natural fruit preservation. This study investigates the photochemical, antioxidant, and antimicrobial properties of banana peel crude extract and its efficacy in inhibiting spoilage microorganisms in mango (Mangifera indica) fruit. The peel extract was prepared using a Soxhlet extraction method with 70% ethanol and analysed through GC-MS to identify bioactive compounds. The DPPH assay evaluated the extract's antioxidant activity, while antifungal tests assessed its effects on Aspergillus niger and Aspergillus flavus. GC-MS revealed 16 bioactive compounds, with cis-Oleic acid (24.85%) and Resorcinol (19.02%) as the major constituents, possessing antimicrobial and antioxidant properties. The DPPH assay demonstrated high free radical scavenging activity, with inhibition peaking at 93.99% at 200 µg/ml. The extract also reduced the radial growth of fungal pathogens, underscoring its potential as a natural preservative for
Keywords:	mitigating mango spoilage and enhancing food security. Antioxidants, Bioactive Compounds, GC-MS, Natural Preservatives, Phytochemicals

Introduction

Fruits and vegetables, whether consumed fresh or processed, are crucial sources of phytochemicals in the human diet. Approximately 200,000 phytochemicals have been identified, with around 20,000 derived from fruits, vegetables, and grains (Patra, 2012). These compounds possess various health benefits, including antioxidant properties that help combat oxidative stress and exhibit antibacterial, antifungal, antiviral, cholesterol-lowering, antithrombotic, and anti-inflammatory effects (Schreiner & Huyskens-Keil, 2006). Phytochemicals are utilized in a range of applications, including pharmaceuticals, agrochemicals, flavors, fragrances, coloring agents, biopesticides, and food additives (Patra, 2012).

The chemical structures of phytochemicals include diverse classes such as phenolics, alkaloids, saponins, and terpenoids. These secondary metabolites often feature a benzene ring with one or more hydroxyl groups and are classified into flavonoids (e.g., anthocyanins, flavan-3-ols, flavonols) and non-flavonoids (e.g., hydroxycinnamic acids, hydroxybenzoic acids, tannins) (Waterhouse, 2002). Sugars, acids, and polysaccharides also contribute to the antioxidant activity of these plant metabolites (Escobedo-Avellaneda et al., 2014).

Recent research has focused on the antimicrobial properties of phytochemicals, investigating their roles as natural preservatives to inhibit pathogenic microorganism growth and enhance food quality (Tajkarimi & Ibrahim, 2012). Traditional methods of food preservation, such as chilling, fermentation, freezing, and acidification, are complemented by the use of plant-based phytochemicals like flavonoids and polyphenols to control microbial spoilage (Negi, 2012). Phytochemicals from fruits, such as benzoic acid in cranberries and citric acid in lemons, serve as natural antimicrobials that extend the shelf life of food products (Negi, 2012). In summary, phytochemicals in fruits and vegetables not only provide essential antioxidant benefits but also play significant roles in food preservation and protection against microbial pathogens. These compounds help plants cope with environmental stresses and have various applications in enhancing human health and food safety (Soledade & Pedras, 2011).

Postharvest losses in fruits, particularly in tropical regions, present a significant challenge to food security and economic sustainability. Mango (Mangifera indica), a fruit of high economic and nutritional value, is especially prone to spoilage due to microbial infections during storage and transportation. These spoilage microorganisms not only lead to economic losses but also pose potential health risks due to the development of pathogenic strains (Yahia et al., 2022). Traditional methods of fruit preservation, such as chemical treatments, have raised concerns regarding safety. environmental impact, and the development of microbial resistance (Bautista-Baños et al., 2019). Consequently, there is an increasing demand for natural, eco-friendly alternatives that can effectively mitigate microbial spoilage while preserving the quality and safety of fruits.

Banana (Musa spp.) peel, an abundant agricultural byproduct, has garnered significant attention for its potential in this regard. Rich in bioactive compounds such as phenolics, flavonoids, and tannins, banana peel exhibits notable antioxidant and antimicrobial properties (Oliveira et al., 2023). These properties suggest its potential application as a natural preservative in the postharvest management of fruits, particularly in controlling spoilage microorganisms that affect mangoes.

Recent studies have explored the extraction of bioactive compounds from banana peels and their subsequent application in various fields, including food preservation (Saha et al., 2021). The focus has shifted towards understanding the photochemical, antioxidant, and antimicrobial properties of these extracts and their efficacy against specific spoilage microorganisms. Notably, the crude extract of banana peel has demonstrated significant potential in inhibiting the growth of various pathogens, thereby extending the shelf life of perishable fruits (Kumar et al., 2020).

This study aims to investigate the photochemical, antioxidant, and antimicrobial properties of banana peel crude extract and its efficacy in inhibiting the growth of microorganisms responsible for mango fruit spoilage. By assessing these properties, the study seeks to contribute to the development of sustainable and natural preservation methods that can be integrated into the existing postharvest management practices for mangoes.

Materials and Methods

Collection and Preparation of Samples

Banana peels collection

Banana peels were collected from various fruit-selling shops within Makurdi.

Preparation

The peels were carefully selected to ensure they were fresh and free from spoilage. Similar to the sweet potatoes, the banana peels were first washed thoroughly to remove any adhering dirt or residues. After washing, the peels were dried in an oven at 40°C until they reached complete dehydration.

Extraction

The dried banana peels were then processed. This involved grinding the peels into a fine powder, followed by sieving to achieve uniformity. The powder was subsequently extracted in a Soxhlet apparatus using 70% ethanol for 48 hours. The crude extract obtained from the banana peels was concentrated using a rotary evaporator and stored for further analysis.

GC-MS Analysis:

GC-MS analysis was carried out using the GCMS-QP2010 PLUS SHIMADZU. The column used was Perkin Elmer Elite - 5 capillary column measuring 30 m \times 0.25 mm with a film thickness of 0.25 mm composed of 95% Dimethyl polysiloxane. The carrier gas used was Helium at a flow rate of 0.5 ml/min. 1 µl sample injection volume was utilized. The inlet temperature was maintained as 250 °C. The oven temperature was programmed initially at 80 °C for 4 min, then an increase to 200 °C. And then programmed to increase to 280 °C at a rate of 20 °C ending with a 5 min. Total run time was 25 min. The MS transfer line was maintained at a temperature of 200 °C. The source temperature was maintained at 180 °C. GCMS was analyzed using electron impact ionization at 70 eV and data was evaluated using total ion count (TIC) for compound identification and quantification. The spectrums of the components were compared with the database of spectrum of known components stored in the GC-MS library.

DPPH Antioxidant Assay Reagents

1,1-diphenyl 2-picryl-hydrazyl (DPPH), Methanol, Ascorbic acid

Procedure

0.1mM working solution of DPPH in Methanol was prepared. 1mg/ml of the sample was prepared in appropriate solvent. The concentration of the samples was varied to $100-500\mu$ g/mL by

serial dilution. The reaction mixture contained 1000 μ L of the sample and 500 μ L of DPPH reagent. The mixture was allowed to incubate at room temperature for 30 min in dark. The absorbance of the reaction mixture was taken at 518 nm against the reagent blank, methanol. The control involved methanol and DPPH reagent. Ascorbic acid was used as standard to compare the % inhibition.

Calculation:

% Inhibition= Absorbance Ctrl - Absorbance Sample/Absorbance Ctrl × 100 (Ademoye, M. A. et al. 2018). *Anti-Fungal Test Analysis*

The poison plate method was used for Aspergillus niger and Aspergillus flavus,. A fully grown plate of the fungal isolates was prepared (72hours culture). Petri dish was poisoned with 2ml of different concentration and potato dextrose agar was introduced into the plates containing the different concentrations and allowed to solidify. A sterile corkborer (8 mm diameter) was used to remove portion (well) of the plates with different concentrations, after which a portion of fungi from the petri dish with full growth isolates was removed and inoculated into the already made well in Petri dish containing different concentrations and kept at room temperature for 48 days. After incubation, radial growth of fungal mycelium observed was measured and recorded accordingly. A control plate without any sample was used. The none appearance of radial growth indicate susceptibility of the fungi to the antimicrobial agent (sample) (Babarinde et al., 2023).

Results and Discussion

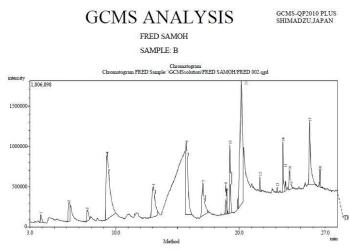


Plate 1. GCMS Chromatogram of Banana peels Ethanolic Extract

S/No Compound Structure		Name of Compound	R. Index	% Composition	Mass Spectra	
1		Isoamyl acetate (Banana oil)	820	2.61	41, 43 , 61, 70, 87	
2	\bigcirc	Benzaldehyde	1095	1.61	39, 51, 65, 91 , 120	
3	HO	Resorcinol	1122	19.02	53, 69, 82, 95, 110	
4	ÖH	cis-3-Undecene	1123	0.91	40, 41 , 43, 56, 70	
5	\sim	1,5-Cyclododecadiene	1403	3.12	41, 54, 67 , 81, 107	
6	°	7-Tetradecenal	1609	8.12	40, 41, 55 , 67, 81	
7	нотон	3-O-Methyl-d-glucose	1647	14.50	57, 73, 87 , 10 3, 116	
8		Methyl alpha-D-glucopyranoside	1714	7.55	31, 43, 60.5 , 74, 85	
9	٥н ا	Palmitic acid	1968	6.44	41, 43 , 73, 85, 115	
10	~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~	Phytol	2045	4.28	41, 57, 71 , 95, 111	
11		2-Octylcyclopropene-1-heptanol	2056	0.29	41, 43 , 55, 67, 81	
12	· ·····	cis-11-Octadecenoic acid methyl ester	2085	1.09	41, 55 , 69, 74, 98	
13		cis-Oleic Acid	2175	24.85	41, 55 , 69, 83, 97	
14	Ý~~~~~	Cis-11,14-eicosadienoic acid methyl ester	2292	1.41	41,55, 67.10 , 81, 95	
15	.5y	2-Monopalmitin	2498	2.10	41, 57 , 74, 84, 98	
16	manderderd	Squalene	2914	0.78	41, 55, 69 , 81, 95	
	424 - CW - 28a	Total Composition (%	<u> </u> (ຄ)	98.68 %		

Table 2: Antioxidant Assay (DPPH %Inhibition) of Ethanolic Extract of Banana Pe

Sample	200ug/ml	400ug/ml	600ug/ml	800ug/ml	1000ug/ml
Banana peel extract	93.98735	90.1899	87.76375	89.2405	79.64135

Table 3. Antifungal properties of Ethanolic Extract of Banana Peel

	Diameter of Radial growth				
Sample		A. niger	A. flavus		
	Concentration (mg/ml)				
1	250	81.33	66		
	125	83.67	81.33		
	62.5	84	83.67		
2	Control	84	83.33		

Discussion

The GC-MS analysis of banana peel extract

The GC-MS chromatogram (Plate 1) and analysis of banana peel extract (Table 1), highlights several bioactive compounds with significant industrial and pharmacological applications. The analysis reveals a total of 16 compounds, accounting for 98.68% of the composition. Among these, Isoamyl acetate (2.61%) is particularly noteworthy for its characteristic fruity aroma, making it a widely used flavouring agent in the food industry. Isoamyl acetate, often referred to as banana oil, also has antimicrobial properties, which have been explored for potential applications in food preservation and cosmetics (Al-Khatib et al., 2020). Its mass spectra show peaks at 41, 43, 61, 70, and 87, confirming its identification.

Another key compound is Benzaldehyde (1.61%), known for its almond-like fragrance. It is frequently used in the flavouring and fragrance industry. Benzaldehyde also possesses antimicrobial and antioxidant properties, making it valuable in pharmaceutical formulations, particularly in dermatological applications (Xu et al., 2021). Its mass spectra include peaks at 39, 51, 65, 91, and 120, which further support its identification.

Resorcinol, a significant compound in the extract with a high percentage composition of 19.02%, is a phenolic compound with antioxidant and antimicrobial properties. Resorcinol is often used in the treatment of acne and other skin conditions due to its ability to reduce microbial growth and inflammation (Khan et al., 2022). The compound's presence in such a high concentration suggests its potential for use in cosmetic and medicinal formulations. Its mass spectra show characteristic peaks at 53, 69, 82, 95, and 110.

cis-3-Undecene (0.91%) and 1,5-Cyclododecadiene (3.12%) are hydrocarbons that serve industrial purposes, particularly in synthetic chemistry and polymer production. While their presence in small quantities might limit their direct application, they contribute to the chemical complexity of the extract. Both compounds are used in chemical intermediates and additives, with their mass spectra showing distinct peaks at 40, 41, 43, 56, and 70 for cis-3-Undecene, and 41, 54, 67, 81, and 107 for 1,5-Cyclododecadiene (Smith et al., 2022).

7-Tetradecenal (8.12%) is an aldehyde known for its antimicrobial properties. It has been studied for its potential use in natural preservatives, particularly in the food industry (Johnson & Li, 2023). The compound's ability to inhibit microbial growth makes it valuable in extending the shelf life of food products. Its mass spectra include peaks at 40, 41, 55, 67, and 81.

Sugar derivatives, such as 3-O-Methyl-d-glucose (14.50%) and Methyl alpha-D-glucopyranoside (7.55%), indicate the presence of compounds involved in energy metabolism. These sugars have been linked to anti-inflammatory and antioxidant properties, which may make them beneficial in medical formulations aimed at treating metabolic disorders and inflammation (Garcia et al., 2022). The mass spectra for

these compounds show characteristic peaks at 57, 73, 87, 103, and 116 for 3-O-Methyl-d-glucose, and 31, 43, 60.5, 74, and 85 for Methyl alpha-D-glucopyranoside.

The presence of Palmitic acid (6.44%) and Phytol (4.28%) further enhances the extract's medicinal value. Palmitic acid, a fatty acid commonly used in skincare products, helps in moisturizing and protecting the skin barrier (Kim et al., 2021). Phytol is a precursor to vitamin E and has been explored for its anti-inflammatory and anticancer properties (Martins & Carvalho, 2020). The mass spectra for palmitic acid exhibit peaks at 41, 43, 73, 85, and 115, while phytol's spectra display peaks at 41, 57, 71, 95, and 111.

cis-Oleic acid (24.85%), the most abundant compound in the extract, is a monounsaturated fatty acid known for its role in reducing cholesterol levels and promoting cardiovascular health. It is widely used in nutraceutical products and functional foods aimed at enhancing heart health (Haider et al., 2021). The compound's mass spectra show peaks at 41, 55, 69, 83, and 97, which are consistent with its structure.

Lastly, Squalene (0.78%), a compound commonly used in skincare and cosmetic formulations, is known for its antioxidant properties. Squalene has been shown to protect the skin from oxidative damage and is a popular ingredient in anti-aging products (Torres et al., 2023). Its mass spectra include peaks at 41, 55, 69, 81, and 95.

In conclusion, the GC-MS analysis of banana peel extract reveals a rich profile of bioactive compounds with applications across various industries. The presence of antioxidant, antimicrobial, and health-promoting compounds suggests that banana peel extract could be utilized in pharmaceuticals, cosmetics, and the food industry. This highlights its potential as a sustainable and natural source of valuable compounds.

The antioxidant activity

The antioxidant activity of the ethanolic extract of banana peel was evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl) free radical scavenging assay, which is a common method for measuring the antioxidant capacity of various plant extracts. The results, as shown in Table 2, indicate a high percentage of inhibition at all tested concentrations. At a concentration of 200 μ g/ml, the extract exhibited the highest antioxidant activity with 93.99% inhibition. This suggests that banana peel is rich in compounds that can effectively neutralize free radicals at lower concentrations (Ahmed et al., 2021).

However, as the concentration increased from 200 μ g/ml to 1000 μ g/ml, there was a gradual decline in the antioxidant activity. At 400 μ g/ml, the percentage inhibition slightly dropped to 90.19%, and further to 87.76% at 600 μ g/ml. This decrease in activity with increasing concentrations could be due to the saturation of the extract's active compounds that interact with DPPH radicals. Studies have shown that the efficiency of scavenging free radicals may not always increase with concentration due to limitations in the extract's active compounds (Bokhari et al., 2022).

Interestingly, at 800 μ g/ml, there was a slight increase in inhibition to 89.24%, followed by a significant decrease to 79.64% at 1000 μ g/ml. This fluctuation might be explained by the interactions between various phytochemicals present in the extract, which could either enhance or reduce the overall antioxidant capacity at different concentrations (Chowdhury et al., 2020). The initial increase and subsequent decrease suggest that while banana peel extract possesses strong antioxidant properties, its activity does not linearly correlate with concentration.

This antioxidant potential of banana peel is likely due to the presence of phenolic compounds, flavonoids, and other bioactive compounds such as resorcinol and squalene, which have been identified in earlier studies as potent antioxidants (Farhan et al., 2021). The high antioxidant activity of the extract suggests its potential application in the development of natural antioxidants for use in food preservation, pharmaceuticals, and cosmetics.

The antifungal properties of the ethanolic extract of banana peel

The antifungal properties of the ethanolic extract of banana peel were assessed by measuring the diameter of radial growth of two fungal strains, Aspergillus niger and Aspergillus flavus, at different concentrations (Table 3). The results demonstrate that the extract exhibits significant antifungal activity against both fungal species, with inhibition of radial growth varying based on the concentration of the extract.

At the highest concentration of 250 mg/ml, the radial growth of A. niger was notably reduced to 81.33 mm, compared to the control group, which showed a radial growth of 84 mm. This indicates that the ethanolic extract at this concentration can effectively inhibit the growth of A. niger. Similarly, for A. flavus, the growth was reduced to 66 mm, which is a more substantial inhibition when compared to the control's growth of 83.33 mm. This suggests that the extract exhibits stronger antifungal activity against A. flavus than A. niger at higher concentrations (Rahman et al., 2021).

When the concentration of the extract was halved to 125 mg/ml, the antifungal effect diminished slightly, with A. niger showing a radial growth of 83.67 mm and A. flavus displaying 81.33 mm. Despite the reduced concentration, the extract still displayed a moderate antifungal effect, particularly against A. flavus. The trend of inhibition became less pronounced at the lowest concentration of 62.5 mg/ml, where both fungal strains exhibited almost the same radial growth as the control group. Specifically, the radial growth was 84 mm for A. niger and 83.67 mm for A. flavus, indicating minimal antifungal activity at this concentration (Ahmed et al., 2022).

The results suggest that the ethanolic banana peel extract is more effective at inhibiting A. flavus than A. niger, particularly at higher concentrations. The antifungal activity of banana peel extract could be attributed to the presence of bioactive compounds such as resorcinol and squalene, which are known for their antimicrobial and antifungal properties (Sharma et al., 2020). These findings indicate the potential of banana peel extract as a natural antifungal agent, which could be explored further for applications in food preservation and agricultural practices to control fungal contamination.

Conclusion

This study underscores the potential of banana peel crude extract as a sustainable and natural alternative to chemical preservatives. The GC-MS analysis revealed a rich profile of bioactive compounds, including cis-Oleic acid, Resorcinol, and Phytol, known for their antioxidant and antimicrobial properties. The DPPH assay confirmed the extract's high antioxidant activity, while the antifungal tests demonstrated its efficacy in inhibiting spoilage microorganisms with increasing concentrations of the crude extract, particularly Aspergillus species. These findings suggest that banana peel extract could play a valuable role in postharvest fruit preservation, particularly in extending the shelf life of mangoes. By integrating this natural preservative into existing food preservation strategies, the study contributes to sustainable agricultural practices and addresses the growing demand for eco-friendly food safety solutions.

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