COMPARATIVE ANALYSIS OF PHYTOCHEMICAL COMPOSITION, PROXIMATE CONTENT, AND HEAVY METAL CONTAMINATION IN NATURALLY AND CALCIUM CARBIDE RIPENED BANANAS

ABSTRACT

The use of artificial ripening agents, particularly calcium carbide, in fruits like bananas has raised concerns about alterations in their nutritional content. This study aimed to compare the phytochemical composition, proximate analysis, and heavy metal contamination in naturally ripened bananas and calcium carbide-ripened bananas. Mature, unripe bananas were purchased and divided into two groups. One group ~~to ripen~~ripened naturally at room temperature, while the other was artificially ripened using calcium carbide. Phytochemical analysis revealed the presence of phenols, alkaloids, flavonoids, tannins, saponins, and terpenoids in both banana types, with significant variations in concentration. Cyanogenic glycoside and oxalate levels were higher in calcium carbide-ripened bananas (7.0 x 10-4), and flavonoid, alkaloid, and saponin concentrations were higher (11.64%, 4.50%, and 9.84%) in naturally ripened fruits, suggesting a reduction in antioxidant properties. Proximate analysis showed higher moisture content in the calcium carbide-ripened bananas (82.05%) compared to naturally ripened bananas (79.83%), while naturally ripened bananas contained higher protein (1.88%), carbohydrate (19.68%), and fiber (2.40%). Heavy metal analysis detected higher phosphorus levels (184.5) in calcium carbide-ripened bananas~~ (184.5)~~, which exceeded the FAO's permissible limits of 80–120 ppm. The study underscores the importance of regulating the use of artificial ripening agents in fruits to safeguard consumer health.

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It is also very necessary to evaluate the effect of this treatments (naturally ripened and calcium carbide ripened) banana on the dominant micronutrient composition (Vitamins and minerals) of this banana samples

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Keywords: ~~calcium ~~Calcium carbide;~~,~~ ~~banana~~Banana;~~,~~ ~~phytochemical ~~Phytochemical analysis;~~,~~ ~~proximate ~~Proximate analysis;~~,~~ ~~heavy ~~Heavy metals;~~,~~ ~~artificial ~~Artificial ripening;~~,~~ ~~food ~~Food safety.

Introduction

Bananas (*Musa spp.*) are one of the most popular and widely consumed fruits globally, recognized for their rich nutritional content and health benefits. They are a significant source of vitamins, minerals, dietary fibre, and bioactive compounds, making them a staple in many diets around the world (Annor et al., 2016). In many developing countries, bananas play a crucial role in food security and are consumed either raw or as processed products. However, due to their perishable nature, bananas are often subjected to artificial ripening processes to meet consumer demand and extend their market availability.

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Calcium carbide, a common chemical agent used for artificial ripening, has raised significant health and safety concerns. When calcium carbide encounters moisture, it produces acetylene gas, which accelerates the ripening process. This method, while efficient, may introduce harmful substances into the fruit, such as phosphine, and negatively impact the fruit's nutritional composition (Islamiyat et al., 2016). Several studies have highlighted the adverse effects of calcium carbide on the quality and safety of ripened fruits, including potential toxicities and nutrient degradation (Asif, 2012; Igbinaduwa et al., 2022). Despite these risks, calcium carbide remains widely used in many countries, including Nigeria, due to a lack of regulatory enforcement and consumer awareness.

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The nutritional and phytochemical profile of fruits, including bananas, is largely influenced by the ripening process. Naturally ripened bananas undergo a series of biochemical changes that enhance their sweetness, flavor, and nutritional value. Conversely, artificially ripened fruits may experience an altered balance of bioactive compounds, potentially affecting their health benefits. Phytochemicals, such as phenols, flavonoids, and alkaloids, are particularly sensitive to the ripening process and play a crucial role in antioxidant defense and disease prevention (Izundu et al., 2016).

Given the widespread use of calcium carbide as a ripening agent and the growing concerns about its safety, this study aims to compare the phytochemical composition, proximate analysis, and heavy metal content of naturally ripened and calcium carbide-ripened banana fruits. By

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METHODOLOGY

SAMPLE COLLECTION AND PREPARATION

Banana Acquisition: Matured unripe bananas were purchased from the Institute of Agricultural Research and Development (IARD). The fruits were divided into two equal parts: one was left to ripen naturally at room temperature, while the other was artificially ripened using calcium carbide. Specifically, 5 g of calcium carbide was used to ripen 1 kg of bananas, following the method of Igbinaduwa et al. (2018).

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Sample Processing: Selected ripe banana samples (both naturally and artificially ripened) were washed with distilled water, sliced, sun-dried, ~~ground into powder~~milled using a mortar and pestle, and then sieved through a 2 mm sieve. The powdered samples were stored in airtight containers until further analysis.

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PHYTOCHEMICAL ANALYSIS

Terpenoids: About 5 ml of each extract was mixed with 2 ml of chloroform and 3 ml of concentrated H_2SO_4. A reddish-brown coloration indicated the presence of terpenoids (Ejikeme et al., 2014).

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Steroids: A mixture of 2 ml of acetic anhydride and 0.5 g of ethanol extract was treated with 2 ml of H_2SO_4, changing color from violet to blue/green to indicate the presence of steroids.

Alkaloids: Alkaloids were extracted by weighing 5 g of the dried sample into a 250 ml beaker, adding 200 ml of 10% acetic acid in ethanol, and allowing it to stand for 4 hours before filtration and precipitation with concentrated ammonium hydroxide (Harborne, 1973).

Saponins: A 20 g sample was extracted with 100 ml of 20% aqueous ethanol, and heated for 4 hours, and the combined extract was purified with n-butanol (Obadoni and Ochuko, 2001).

Flavonoids: Extract 10 g of the sample with 100 ml of 80% aqueous methanol, filter, and evaporate to dryness (Sufowara, 1993).

Tannins: Extraction involved boiling 0.1 g of the sample in water, followed by color development with Folin-Denis reagent (Ejikeme et al., 2014).

Phenols: The sample was boiled in distilled water, treated with NaOH, and titrated with $Na_2S_2O_2$.

Cyanogenic Compounds: 5 g of sample was allowed to hydrolyze overnight before distillation and titration with silver nitrate (Hikino et al., 1984).

Oxalates: 25 cm^3 of the solution was titrated with potassium permanganate after heating with dilute sulfuric acid (Ejikeme et al., 2014).

Phytate Acid: A sample was extracted with HCl, neutralized, treated with $FeCl_3$, and analyzed calorimetrically.

PROXIMATE ANALYSIS

Carbohydrate: 0.1 g of the sample was treated with perchloric acid, and the resulting solution was analyzed using an Anthrone reagent.

Protein: Digestion involved concentrated sulfuric acid, followed by distillation and titration to determine nitrogen content.

Moisture: 1 g of sample was dried at 105°C, and moisture percentage was calculated.

Lipid: Lipids were extracted using a Soxhlet extractor with acetone.

Ash: The sample was incinerated in a muffle furnace, and ash content was calculated.

Crude Fiber: The sample was subjected to a series of extractions and weight measurements to determine fibre content.

HEAVY METAL ANALYSIS

Arsenic and Phosphorus: 5 g of dried sample was ashed at 500°C, dissolved in HNO_3, and analyzed using Atomic Absorption Spectrophotometry (Miroslav and Vladimir, 1999).

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STATISTICAL ANALYSIS

Data were analyzed using one-way Analysis of Variance (ANOVA) with post-hoc LSD tests, considering results significant at $p < 0.05$

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RESULTS

Table 1. Phytochemical Analysis of natural ripened and calcium carbide ripened banana fruit

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	% Cyanogenic glycoside	% Phenols	% Phylate	% Oxalate `	% Alkaloids	% Flavonoid	% Tannin	% Saponin	% Terpenoid
CR	7.0 x 10-4	0.094	1.0 x 10^{-4}	1.82	4.2	10.22	7.5 x 10^{-4}	6.41	+++
NR	6.0 x 10-4	0.094	1.0 x 10^{-4}	0.50	4.50	11.64	3.16 x 10^{-4}	9.84	+++

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Phytochemical Analysis

The phytochemical analysis of naturally ripened (NR) and calcium carbide-ripened (CR) banana fruits revealed differences in the concentration of several compounds (Table 1). Cyanogenic glycoside levels were higher in calcium carbide-ripened bananas (7.0 x 10^{-4}) compared to naturally ripened bananas (6.0 x 10-4), though the difference was not statistically significant ($p>0.05$). Phenols (0.094%) and phytate (1.0 x 10^{-4}) were consistent in both ripening methods.

However, oxalate and tannin levels were significantly elevated in calcium carbide-ripened bananas (1.82) and (7.5 x 10^{-4}) respectively) compared to the naturally ripened ones (0.50) and (3.16 x 10-4). Flavonoid, Alkaloid, and saponin concentrations were higher in naturally ripened bananas (11.64%, 4.50%, and 9.84%, than in calcium carbide-ripened fruits, where alkaloids and saponin levels dropped to 4.2% and 6.41%, respectively. Terpenoid levels remained unchanged between the two groups.

Table 2. Proximate Analysis of Natural Ripened and ~~calcium carbide ripened banana f~~Calcium Carbide Ripened Banana Fruit

	%	%	%	%	%	%	%
	Moisture	M/crushed	Ash	Lipid	Protein	Carbohydrates	Fibre
CR	82.05	83.03	0.21	1.23	1.61	14.16	0.49
NR	79.83	81.11	0.52	1.65	1.88	19.68	2.40

The proximate analysis showed a notable difference in moisture content, which was slightly higher in calcium carbide-ripened bananas (82.05%) compared to naturally ripened bananas (79.83%) (Table 2). The moisture content of calcium carbide-ripened bananas also translated to a crushed moisture content of 83.03%, compared to 81.11% in naturally ripened bananas.

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Ash content was lower in calcium carbide-ripened bananas (0.21%) than in naturally ripened bananas (0.52%). The lipid content also dropped in calcium carbide-ripened bananas (1.23%) compared to naturally ripened bananas (1.65%). Similarly, protein levels in calcium carbide-ripened bananas (1.61%) were slightly reduced compared to naturally ripened ones (1.88%). Carbohydrate content was lower in calcium carbide-ripened bananas (14.16%) versus naturally ripened bananas (19.68%). Fiber content showed a sharp decline in calcium carbide-ripened bananas (0.49%) compared to naturally ripened fruits (2.40%).

Table 3. Heavy Metal Analysis of natural ripened and calcium carbide ripened banana fruit

Heavy metals	Naturally ripened (mg/kg)	Calcium carbide ripened (mg/kg)	FAO STANDARD (ppm)
Arsenic	Not detectable	Not detectable	0.5 -2.0
Phosphorous	77.19	184.5	80 - 120

The heavy metal analysis indicated that arsenic was undetectable in both naturally ripened and calcium carbide-ripened bananas (Table3). However, there was a significant increase in phosphorus content in calcium carbide-ripened bananas (184.5 mg/kg) compared to naturally ripened bananas (77.19 mg/kg). The phosphorus content in calcium carbide-ripened bananas also exceeded the FAO standard of 80–120 ppm, highlighting the potential risk of heavy metal contamination from artificial ripening methods.

Discussion

Phytochemical Analysis

The phytochemical composition of both naturally ripened and calcium carbide-ripened banana fruits indicates the presence of several bioactive compounds such as phenols, tannins, alkaloids, saponins, flavonoids, and glycosides. These compounds are known for their medicinal properties, which makes bananas a valuable source of health-promoting nutrients (Kasolo et al., 2010). However, significant differences were observed between the two ripening methods.

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For instance, the cyanogenic glycoside content was found to be higher in calcium carbide-ripened bananas. This glycoside is a natural plant toxin, which can release cyanide, potentially causing serious health issues like growth retardation and neurological disorders such as neuropathy, seizures, encephalopathy, Amyotrophic lateral sclerosis (ALS), and dementia (Islamiyat et al., 2016). A similar trend was observed in other studies, which reported an increase in cyanogenic glycosides with fruit ripening (Alagbaoso et al., 2017). While the phenol content remained consistent between the two types of ripened bananas, phenolic compounds are known for their antioxidant properties and their ability to protect against diseases like cancer and cardiovascular conditions (Hollman, 2001).

The oxalate content was notably higher in calcium carbide-ripened bananas, which raises concerns about potential kidney issues due to the formation of calcium oxalate stones Gao et al., 2012). This is significant, as bananas are commonly consumed in large quantities, and the accumulation of oxalates over time could pose a health risk (Gbakon et al., 2018).

Interestingly, the alkaloid and flavonoid contents were higher in naturally ripened bananas. Alkaloids are valuable for their antimicrobial, analgesic, and antiplasmodic properties (Iheagwam et al., 2019), while flavonoids contribute to antioxidant defense mechanisms, helping prevent oxidative stress (Izundu et al., 2016). On the other hand, tannin levels were higher in ~~calcium ~~calcium-carbide-ripened bananas. Tannins have astringent properties and are beneficial for wound healing, but in higher doses, they can negatively affect nutrient absorption (Ugwu et al., 2003; Offor et al., 2015).

The study also found a higher concentration of saponins and terpenoids in naturally ripened bananas, which are known for their cleansing, ~~pesticidal~~pesticide, and anticancer properties

(Gurfinkel & Rao, 2003). These findings suggest that naturally ripened bananas may offer superior health benefits compared to those ripened with calcium carbide.

Proximate Analysis

The proximate composition of calcium carbide-ripened bananas revealed an increased moisture content (82.05%) compared to naturally ripened bananas (79.83%). This observation aligns with findings from Nahu et al. (2020), where the moisture content increased with the use of calcium carbide. The elevated moisture content is a result of weakened banana peel fiber, which allows for higher water absorption during ripening. Ash content, which reflects the inorganic mineral content, was found to be lower in calcium carbide-ripened bananas (0.21%) compared to naturally ripened bananas (0.52%), but both fell within the FAO's permissible limits (2.9%). Similar trends were reported by Nahu et al. (2020), where calcium carbide affected mineral absorption in banana peels, although the mineral content remained within safe levels.

The percentage of lipid content was also found to be lower in calcium carbide-ripened bananas, differing from reports by Adeyemi et al. (2018), where calcium carbide increased lipid levels. Protein content was significantly reduced in calcium carbide-ripened bananas, suggesting that calcium carbide may impair the nutritional quality of bananas, particularly their ability to provide essential amino acids and energy (Nura et al., 2018). Carbohydrate content followed a similar pattern, with higher levels in naturally ripened bananas (19.68%) compared to those ripened with calcium carbide (14.16%). This could be attributed to the increased respiration rate in ~~calcium~~ calcium-carbide-treated bananas, which accelerates the conversion of carbohydrates into carbon dioxide (Adeyemi et al., 2018). Fiber content was higher in naturally ripened bananas, which is important for digestive health. Calcium carbide-ripened bananas, on the other hand, showed a decrease in fiber content, which contrasts with other studies, such as Adeyemi et al. (2018), where calcium carbide was associated with increased fibre.

Heavy Metal Analysis

The heavy metal analysis revealed no detectable levels of arsenic in either naturally ripened or calcium carbide-ripened bananas. This aligns with the findings of Igbinaduwa et al. (2018), who reported that laboratory-ripened bananas did not contain arsenic, although market-sourced bananas did. The absence of arsenic in this study suggests that proper handling and control of calcium carbide during ripening may minimize arsenic contamination.

However, phosphorus levels were significantly higher in calcium carbide-ripened bananas (184.5 mg/kg) compared to the naturally ripened ones (77.19 mg/kg), exceeding the FAO standard of 80-120 mg/kg (FAO and WHO, 1998). Prolonged consumption of phosphorus in such high quantities may lead to health issues such as memory loss, mental confusion, and seizures (Fattah & Ali, 2010). This finding is consistent with previous studies that have reported high phosphorus levels in calcium carbide-ripened bananas (Nuhu et al., 2020; Igbinaduwa et al., 2018).

Conclusion

The findings of this study suggest that calcium carbide ripening can significantly alter the phytochemical and proximate composition of bananas, potentially diminishing their nutritional quality and increasing the risk of exposure to harmful substances like cyanogenic glycosides and excess phosphorus. While calcium carbide ripening speeds up the process, it comes at a nutritional and health cost, reinforcing the need for better regulatory measures and public awareness regarding the use of artificial ripening agents.

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Considering the global trend of banning calcium carbide as a ripening agent in countries like India, Sri Lanka, and Bangladesh (Siddiqui & Dhua, 2010), ~~it is imperative for Nigeria to~~Nigeria must enforce similar regulations to safeguard public health.

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