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Research Article

Phytochemical and Antifungal Evaluations of Virgin *Cocos nucifera* (Coconut) Oil

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Abstract

The incidence of antifungal-resistant pathogenic fungi has steadily increased around the globe and calls for an aggressive response. The study aimed to produce and evaluate virgin coconut oil's phytochemical properties and antifungal potentials (VCO). The production of VCO was achieved by hot extraction and natural fermentation methods. Gas Chromatography-Mass Spectrometry (GC-MS) was used to evaluate the active phytochemicals in the VCO. The antifungal activities of VCO against *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans*, *Candida krusei*, *Penicillium chrysogenum*, and *Trichphoyton rubrum* of clinical origin were

determined by the agar-well diffusion method. The percentage yield of VCO obtained from *Cocos nucifera* nuts by hot extraction and natural fermentation methods was 12.80% (w/w) and 11.72% (w/w), respectively. The yield from the hot extraction method was higher but not significantly different from the latter (p = 0.07). The phytochemical analysis revealed twelve chemical compounds. The VCO exerted antifungal effects at various concentrations, except at 3.125 mg/mL. *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans*, and *Penicillium chrysogenum* exhibited resistance to hot-extracted VCO at 6.25 mg/mL. The test organisms were relatively more susceptible to the VCO obtained by natural fermentation than the hot extraction method, but the difference was not statistically significant (p = 0.09). This study revealed that extraction of VCO, with or without heat, possesses antifungal activities and can be used to treat infections caused by pathogenic fungal species. It is beneficial in this era of increasing antimicrobial resistance.

Keywords
Antifungal; antimicrobial resistance; *Cocos nucifera*; fermentation; pathogenic fungi; phytochemicals; virgin coconut oil

1. Introduction

Cocos nucifera (Coconut) is the fruit of the coconut palm, which is used for water, milk, oil, and delicious meat. Coconut trees are native to Southeast Asia and the islands between the Indian and Pacific Oceans and are considered the world's most widely distributed fruit tree [1]. Coconuts are grown globally and are becoming increasingly popular for their flavor, culinary applications, and numerous possible health advantages. Coconut products come in various shapes and sizes; the kernel is the raw white meat within a coconut. It has a firm structure and a tasty, slightly sweet taste [2]. Coconut is commonly found sliced, shaved, or grated in its processed form. The raw, grated meat is pressed to make coconut milk and cream [3]. Grated dried coconut meat is usually used for baking and cooking. It can be ground into flour after further processing [4]. Coconut oil is derived from beef and can be substituted for other vegetable oils in cooking [5].

Fresh coconut milk, flesh, or residue can all be used to make virgin coconut oil (VCO). It is produced from fresh beef by either wet-milling or drying the residue and extracting the oil with a screw press. VCO can also be extracted from fresh meat by shredding and drying it to a moisture content of 10-12%, then pressing the oil out manually. VCO can be used for cooking, fighting infections, removing stains, and removing makeup. It has anti-inflammatory properties, assists digestion, improves metabolism, and lowers arthritis.

Coconut has a variety of practical and medical uses. Coconut is high in minerals, which are engaged in various bodily functions. Coconuts contain high levels of manganese, essential for metabolizing carbohydrates, proteins, cholesterol, and bone health. Coconut meat's high fiber content can aid slow digestion and may even reduce insulin resistance, which can help regulate blood sugar levels [4]. Shredded coconut is fantastic in cookies, muffins, and quick bread because it adds natural sweetness and moisture. Oatmeal with a sprinkle of raw coconut adds texture and a tropical flavor. It is also a tasty calorie booster when mixed with pudding or yogurt for someone

looking to gain weight. In baking, coconut flour is used to replace wheat flour. It is gluten-free, nut-free, and a popular choice for carb-conscious people. The flour is also suitable for those following the paleo diet, which excludes grain items such as regular wheat flour [2].

At 30°C and above, coconut oil is a colorless liquid. It will solidify at a temperature of 25°C. The color of solidified coconut oil is white. The smoking point of unrefined coconut oil is 170°C, while the smoking point of refined coconut oil is 232°C. Only unrefined, bleached, or deodorized coconut oil has a distinctive coconut aroma. Coconut oil is obtained from the coconut fruit and contains capric acid, acid, and lauric acid, which are medium-chain fatty acids. It is utilized in cosmetics, detergent production, and as a culinary oil. Numerous health experts advise restricting its usage as a food due to its high saturated fat content [6].

The incidence of opportunistic fungal infections has steadily increased around the globe, and they are mainly a public health threat in Nigeria and some other African countries. The management of fungal infections (mycoses) is based on using antifungals. However, their use is usually accompanied by some side effects. Moreover, the development of new pathogenic strains and the increased resistance to antifungal agents pose the risk of infection and predispose patients to life-threatening relapses. Antibiotics, including antifungal medicines, have been linked to antifungal resistance in fungi and could manifest in various ways. Antibiotics, for example, can suppress healthy and harmful gut bacteria, allowing opportunistic fungi to thrive [7]. Like other living organisms, fungi can adapt to toxic substances, developing resistance over time. Antifungal resistance refers to a stable, inheritable modification in fungal cells, leading to reduced sensitivity to antifungal agents. This adaptation allows fungal cells to survive exposure to drugs that inhibit or kill them. This calls for researching new therapeutic approaches that can be used as alternatives to conventional therapies. One strategy is using virgin coconut oil, the medicinal potential of which is often underestimated. This study aimed to locally produce virgin *C. nucifera* oil and assess its antifungal and phytochemical properties.

2. Materials and Methods

2.1 Sample Collection

Fifty-three (53) healthy *C. nucifera* nuts were purchased from Odosida and Sabo markets in Ondo City, Nigeria. These were transported in a black polythene bag to the laboratory for analysis.

2.2 Production of VCO

The hot extraction and fermentation methods were employed in this study.

2.2.1 Hot Extraction Process

The coconut was broken to separate the husk from the coconut. The separated coconut was washed thoroughly in warm water and grated. This was soaked for 42 hrs and the coconut milk strained out afterwards. The coconut oil was extracted from coconut milk by heating. The proteins of coconut milk were denatured by the heating process, which destabilized the milk emulsion. The VCO was extracted by heating coconut milk at 100 to 120°C for 60 minutes until the water evaporated completely [8]. To extract the VCO from coconut milk, the protein was coagulated by

gentle heating in the VCO cooker, and the oil formed and was separated from pertinacious residue by filtering through a muslin cloth. The remaining residue was further heated for more oil extraction.

2.2.2 Fermentation Method

Fermentation is a famous cold-process method for extracting VCO from coconut milk [9]. To separate the mature coconut's outer husk from the white edible section, the mature coconut was first carefully broken to separate it from the husk. The coconut was sliced and properly washed with warm water. The sliced nuts were blended with a blender (blender) and transferred into a spotless bucket. About 500 mL of distilled water was added, and the coconut milk was squeezed out of the coconut shaft using a mesh towel. The mixture was then placed into a spotless, clear bucket and covered firmly. The natural fermentation process began as the coconut milk was allowed to settle in the plastic bucket placed in a warm environment and examined after 48 hours. Five layers were formed in the fermentation bucket: the uppermost layer was the fermented curd, followed by separated VCO, fermented curd, fermented skim milk, and the gummy sediment [5].

2.2.3 Getting a Pure Oil

An indigenous technique was employed to obtain pure oil. The oil obtained through this method is commonly known as virgin coconut oil, as it is colorless and does not undergo any heat process. To create a funnel, sterile large bottles were cut in half and stacked on top of one another. The open ends of the bottles were filled with cotton wool, and the upper opening was covered with a mesh cloth. To prevent other materials from mixing with the pure VCO, mesh cloth and cotton wool were used, and a spoon was used to scoop the oil and delicately pour it into the bottles. Following this process, the oil was gathered, placed into sterile bottles, and stored at room temperature.

The percentage yield of VCO obtained from the *C. nucifera* nuts was determined by the formula:

$$\text{Yield} = \frac{\text{Mass of VCO (g)} \times 100}{\text{Mass of the coconut}}$$

2.3 Phytochemistry of VCO

2.3.1 Preparation of the Extract for Phytochemical Screening

The Soxhlet extraction method was used to remove the solvent. The coconut was broken, the husk was taken off, and it was thoroughly cleaned. To let the organic solvent in, the sample was dried to remove the moisture. A more significant exposed surface results from size reduction, which is done to enhance surface area. After that, acidic hydrolysis is carried out, and distillation is used to collect the solvent. After being dried in an oven at 102°C and cleaned with petroleum ether, the glass apparatus was removed and maintained in a desiccator. The thimble containing the 5 g of ground material was weighed and stored in the Soxhlet extractor and the Soxhlet. A round-bottomed container was filled with a 50/50 v/v combination of n-hexane and methanol flask. The condenser was coupled to the extractor, which was connected to a conduit where water was continuously flowing, and the flask was attached to the central Soxhlet extractor. The entire procedure was allowed to boil once the hole setup was placed on a heating mantle. The following eight hours were spent on this process. The extraction unit's condensing unit was removed, and the

sample was left to cool. After distillation, the sample was collected and put in the oven after being removed from the desiccator. The sample's weight was recorded. The cleaned extract was then reduced in volume using nitrogen concentration to about 1 mL before being added to the GC-MS analyzer [10].

2.3.2 Gas Chromatography-Mass Spectrometry (GC-MS) Analysis

The GC-MS analysis was done at the Science and Technology Laboratory, opposite IITA, Moniya, Ibadan, Oyo state, Nigeria. A GC-MS (Modal; Agilent technologies 7890A) fitted with a VF - 5 ms fused silica capillary column of 30 m length, 0.25 mm diameter, and 0.25 mm film thickness was utilized for the gas chromatography-mass spectrometry (GC-MS) examination of VCO. An electron ionization device with an ionization energy of 70 eV was employed for GC-MS detection. As a carrier gas, helium gas (99.99%) was employed at a constant flow rate of 1 mL/min. Temperatures were chosen for the injection and mass transfer lines at 200 and 240°C, respectively. The oven temperature was between 80°C for 2 minutes at 10°C/min and 240°C for 6 minutes. Manually inserting 2 mL of water solution from the samples in split-less mode with a split ratio of 1:40 and a mass scan of 50-600 amu. The GC-MS ran for 35 minutes in total. Peak area normalization was used to express each extract constituent's relative percentage as a percentage. The National Institute of Standard and Technology (NIST) library's database, which has more than 62,000 spectral patterns, was used to interpret the mass spectrum of the extracts. Compound spectra were compared to library database spectra from the National Institute of Standards and Technology (NIST) [10]. By comparing the phytochemicals' binding affinities to the receptor, the phytochemicals extracted were examined.

2.4 Antifungal Analysis of VCO

2.4.1 Collection of Test Organisms

The pure clinical isolates of *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans*, *Candida krusei*, *Penicillium chrysogenum*, and *Trichphoyton rubrum* were obtained from the Department of Microbiology and the Department of Microbial pathology of the University of Medical Sciences, Ondo City, Nigeria.

2.4.2 Antifungal Assay

The antifungal effect of the different concentrations of VCO against the test organisms was evaluated using the agar-well diffusion method. The test organisms were subcultured on potato dextrose agar (PDA) medium to obtain a young actively growing culture. Suspension of the fungal cells equivalent to McFarland standard were prepared to obtain a final inoculum size of 0.5×10^8. A 1-mL portion of the fungal suspension was inoculated on a PDA medium using the spread plate method. Six millimeter-diameter wells were bored on PDA plates, and 0.5 mL of the different concentrations of VCO were introduced separately. These plates stood for 30 mins and incubated at 25°C for 72 h. All experiments were performed in triplicates under aseptic conditions. The antifungal activity was shown by clear zones of inhibition (mm) around the wells, and the average values were calculated and recorded [11]. Uninoculated PDA plates served as negative controls while inoculated well with fluconazole (100 mg/mL) served as positive controls.

2.4.3 Determination of Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC)

The broth dilution method was employed to determine the MIC of the VCO. Six different concentrations (100, 50, 25, 12.5, 6.25, and 3.125 mg/mL) of the VCO were investigated. Test tubes containing a 9-mL portion of potato dextrose broth were prepared and 0.5 mL of the standard inoculum (0.5×10^8) from the fungal suspension, equal to the McFarland standard, was inoculated in each broth. The different concentrations of the VCO were introduced into the tubes and incubated at 25°C for 72 h. The growth in the tubes indicated the turbidity of the medium. The MICs were taken as the lowest concentrations of the VCO that inhibited the growth of the test organisms. The MFCs were determined by subculturing 2 µL from each tube showing no growth into PDA plates and further incubated at 28°C for 72 h. The lowest concentration with no visible growth was defined as MFC indicating 99.5% killing of the original inoculum. All experiments were performed in duplicate and repeated three times [11].

2.5 Statistical Analysis

The investigations were carried out in triplicates, and the descriptive data analyses were determined using Microsoft Excel 2016. The student t-test and one-way analysis of variance (ANOVA) were used to compare the activities of hot-extracted and naturally-fermented VCO and control antibiotics on the test organisms employing IBM SPSS 2020.

3. Results

3.1 Production of VCO

Figure 1 shows the key production stages of virgin coconut oil by hot extraction process. In contrast, the stages involved in the natural fermentation of coconut milk leading to the production of coconut oil are given in Figure 2. The percentage yield of VCO obtained from *C. nucifera* nuts by hot extraction and natural fermentation methods (Table 1) was 12.80% (w/w) and 11.72% (w/w), respectively. The yield from the hot extraction method was higher but not significantly different from that of natural fermentation ($p = 0.07$).

Figure 1 Production of VCO by hot extraction process.

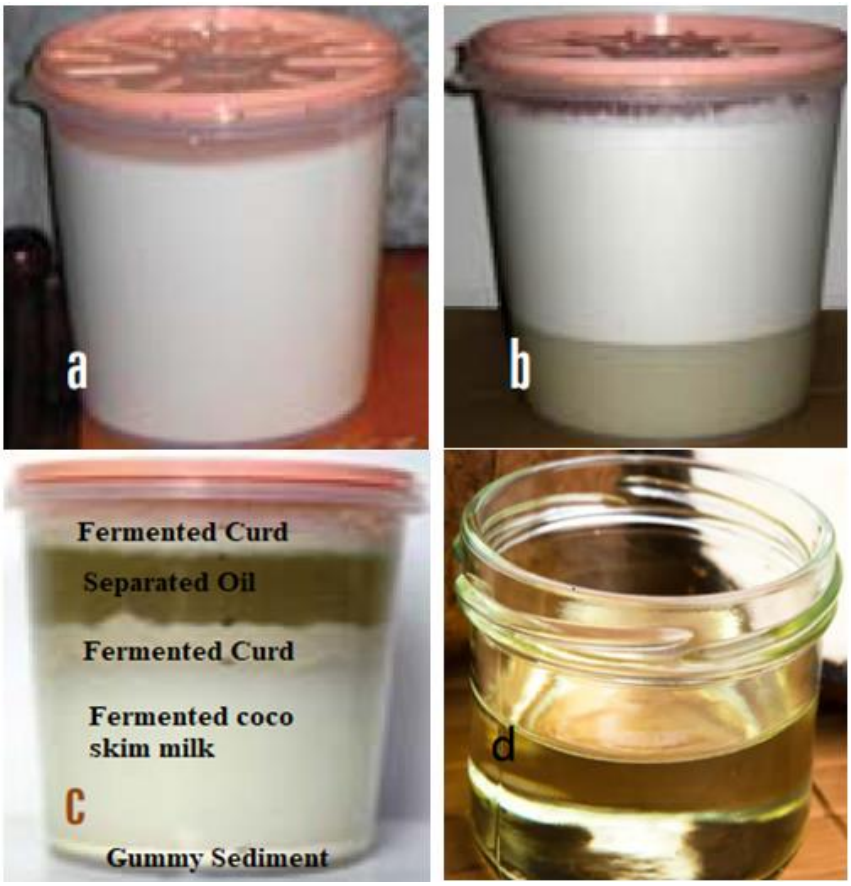


Figure 2 Natural fermentation of coconut milk: initial stage (a), transition stage (b), final stage (c), and separated oil (d).

Table 1 The percentage yield of VCO from *Cocos nucifera* nuts with hot extraction and natural fermentation methods.

Cocos nucifera	Measurement	
	Hot Extraction	Natural Fermentation
Number of coconuts processed	25	25
Weight of the 25 coconuts (g)	3,602[a]	3,625[a]
Weight of the edible part of the nuts (g)	2,231[a]	2,250[a]
Weight of the coconut milk (g)	1,269[a]	1,237[a]
Volume of the coconut milk (mL)	1,306[a]	1,355[a]
Weight of the virgin coconut oil (g)	461[a]	425[a]
Volume of the virgin coconut oil (mL)	448[a]	403[a]

The statistical difference between the yields from both methods was determined at $\alpha \leq 0.05$. Values with the same superscript along the same row showed no statistical difference.

3.2 Phytochemistry of VCO

The gas chromatogram revealed 12 peaks of the compounds present in the VCO (Figure 3). The 12 chemical compounds in the VCO were neophytadiene; 2-pentadecanone 6,10,14-trimethyl; hexadecanoic acid, methyl ester; 9,12-octadecadienoic acid methyl ester; 9,12,15-octadecatrienoic acid, methyl ester; 3,7,11,15-tetramethyl-2-hexadecene-1-ol; methyl stearate; squalene; methyltris(tetramethylsiloxy)silane; cyclotrisiloxane hexamethyl-; 1,1,1,3,5,5,5-heptamethyltrisiloxane; and beta and alpha amyrin. The values of the peak, retention time, peak area, molecular formula, molecular weight, chemical structure, and names of the compounds are given in Table 2.

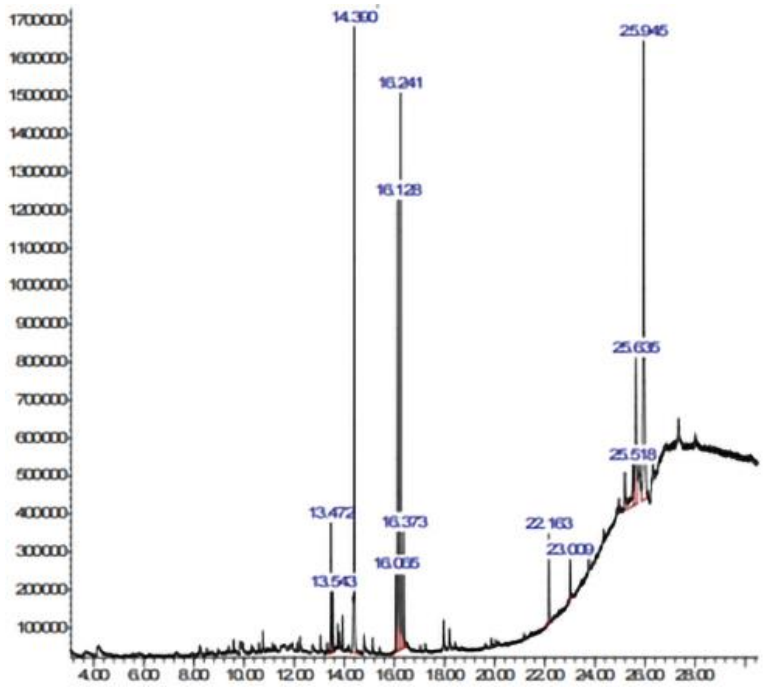
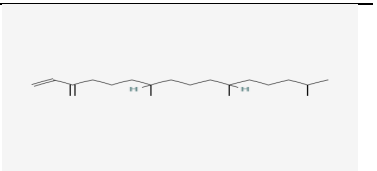
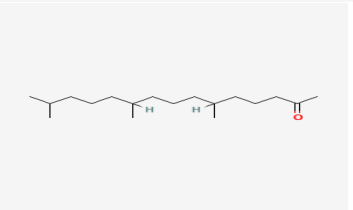
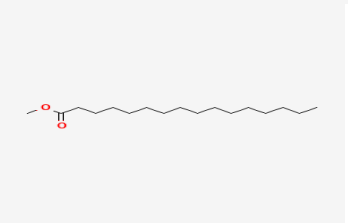
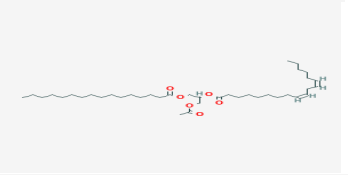
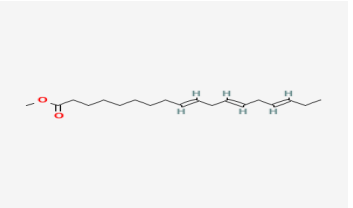
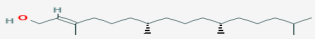
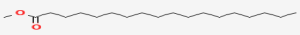
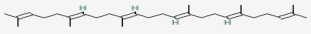
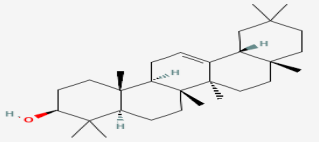
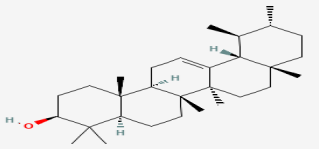


Figure 3 A gas chromatogram showing 12 peaks depicting compounds in *Cocos nucifera* extract.

Table 2 Phytocomponents of *Cocos nucifera* oil extracts using GC-MS analysis.

Peak #	Retention time	Peak area	Name of compound	Molecular formula	Molecular weight	Chemical structure
1	13.472	3.12	Neophytadiene	$C_{20}H_{38}$	278.5	
2	13.541	1.70	2-pentadecanone,6,10,14-trimethyl	$C_{18}H_{36}O$	268.5	
3	14.388	18.54	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270.5	
4	16.064	2.26	9,12,-Octadecadienoic acid methyl ester	$C_{39}H_{70}O_6$	635.0	
5	16.127	14.11	9,12,15-Octadecatrienoic acid, methyl ester	$C_{19}H_{32}O_2$	292.5	

6	16.242	16.55	3,7,11,15-tetramethyl-2-hexadecene-1-ol	$C_{20}H_{40}O$	296.5	
7	16.373	3.75	Methyl stearate	$C_{19}H_{38}O_2$	298.5	
8	22.164	2.83	Squalene	$C_{30}H_{50}$	410.7	
9	23.011	1.32	Methyl tris(tetramethylsilane)silane OR Tetrasiloxane, dimethyl-	$C_{10}H_{30}O_3Si_4$	310.68	
10	25.517	3.52	Cyclotrisiloxane, hexamethyl-	$C_6H_{18}O_3Si_3$	222.46	
11	25.637	8.16	1,1,1,3,5,5,5-heptamethyltrisiloxane	$C_7H_{21}O_2Si_3$	221.50	

			.beta.-Amyrin	$C_{30}H_{50}O$	426.7	
12	25.946	24.15				
			.alpha.-Amyrin	$C_{30}H_{50}O$	426.7	

3.3 Antifungal Assay

The antifungal activities of VCO obtained by the hot extraction and natural fermentation methods exerted inhibitory effects on the test organisms at various concentrations, except at 3.125 mg/mL (Table 3). *Aspergillus flavus*, *Aspergillus niger*, *Candida albicans*, and *Penicillium chrysogenum* exhibited resistance to hot-extracted VCO at 6.25 mg/mL. The test organisms were relatively more susceptible to the VCO obtained by natural fermentation, especially at lower concentrations, than the hot extraction method, but the difference was not statistically significant ($p = 0.09$). The MIC and MFC of VCO derived by the hot extraction process against the test organisms ranged from 3.125 to 6.25 mg/mL and 12.5 to 25 mg/mL, respectively. For naturally-fermented VCO, the MIC was 3.125 mg/mL and MFC ranged from 12.5 to 25 mg/mL (Table 4).

Table 3 The antifungal effects of VCO against test organisms.

Fungi	Fluconazole (100 mg/mL)	Average diameters of zones of inhibition (mm)											
		Concentrations of hot-extracted VCO (mg/mL)						Concentrations of naturally-fermented VCO (mg/mL)					
		100	50	25	12.5	6.25	3.125	100	50	25	12.5	6.25	3.125
Aspergillus flavus	30.5^{a}	26.0^{a}	21.0^{a}	18.0^{b}	12.5^{b}	0^{c}	0^{c}	28.0^{a}	24.5^{a}	19.5^{b}	14.0^{b}	7.0^{d}	0^{c}
Aspergillus niger	27.0^{a}	26.5^{a}	22.0^{a}	18.0^{b}	14.0^{b}	0^{c}	0^{c}	29.0^{a}	24.0^{a}	19.0^{b}	15.5^{b}	6.5^{d}	0^{c}
Candida albicans	28.0^{a}	25.0^{a}	20.0^{a}	13.5^{b}	12.0^{b}	0^{c}	0^{c}	27.0^{a}	22.5^{a}	16.5^{b}	14.5^{b}	6.0^{d}	0^{c}
Candida krusei	27.5^{a}	23.5^{a}	20.5^{a}	17.5^{b}	15.0^{b}	6.5^{d}	0^{c}	27.0^{a}	25.0^{a}	20.0^{b}	16.0^{b}	8.0^{d}	0^{c}
Penicillium chrysogenum	28.5^{a}	25.5^{a}	19.0^{b}	15.5^{b}	12.0^{b}	0^{c}	0^{c}	27.0^{a}	23.0^{a}	17.0^{b}	14.0^{b}	6.5^{d}	0^{c}
Trichophyton rubrum	30.0^{a}	26.0^{a}	22.5^{a}	20.0^{b}	17.5^{b}	9.0^{d}	0^{c}	27.5^{a}	25.5^{a}	19.0^{b}	16.0^{b}	7.0^{d}	0^{c}

Values show the mean of triplicate samples. Values with the same superscript along the same row show no statistical difference. α was taken to be $\leq$ 0.05.

Table 4 The MIC and MFC of VCO against the test organisms.

Fungi	Hot-extracted VCO (mg/mL)		Naturally-fermented VCO (mg/mL)	
	MIC	MFC	MIC	MFC
Aspergillus flavus	6.25	12.5	3.125	12.5
Aspergillus niger	6.25	25	3.125	25
Candida albicans	3.125	25	3.125	12.5
Candida krusei	3.125	25	3.125	12.5
Penicillium chrysogenum	6.25	25	3.125	12.5
Trichphyton rubrum	3.125	25	3.125	25

4. Discussion

The aroma and color of the produced VCO were consistent with those produced by other methods. Aside from the hot extraction and natural fermentation methods (Figure 1 and Figure 2) employed for the extraction of VCO in this study, Agarwal and Bosco [9] reported that VCO could also be produced by the cold extraction method which is achieved by extracting the coconut oil from coconut milk by breaking the emulsion without heating. The chilling, freezing, and thawing method entails the process where the stability of coconut milk emulsion is broken by chilling, freezing, and thawing, and thawed cream is separated by centrifugation [9, 11]. The centrifugation method involves the extraction of VCO employing various centrifugation speeds, time intervals, and temperature [12, 13], while the aqueous enzymatic extraction method is used for VCO extraction by the use of enzymes in the aqueous extraction process [9]. The percentage yields obtained in this study were 12.80% (hot extraction) and 11.72% (natural fermentation) (Table 1). These were higher than the percentage yield of 9.34% reported by Udensi *et al*. [14].

The result of the phytochemical screening of *C. nucifera* oil showed the presence of 12 compounds: neophytadiene; 2-pentadecanone 6,10,14-trimethyl; hexadecanoic acid, methyl ester; 9,12-octadecadienoic acid methyl ester; 9,12,15-octadecatrienoic acid, methyl ester; 3,7,11,15-tetramethyl-2-hexadecene-1-ol; methyl stearate; squalene; methyltris (tetramethylsiloxy)silane; cyclotrisiloxane hexamethyl-; 1,1,1,3,5,5,5-heptamethyltrisiloxane; and beta and alpha amyrin (Table 2). The findings of this study differ from the results of Obidoa *et al.* [15] who did a phytochemical analysis on the milled endosperm of *C. nucifera* using both non-polar (n-hexane) and polar (water) solvents, and reported the presence of phytochemicals like; flavonoids, alkaloids, saponins, tannins, glycosides, steroids, acidic compounds, terpenoids, and resins. This is also in line with Aina *et al.* [16] who worked on *C. nucifera* oils from two locations in Kaduna state, Nigeria. The presence of alkaloids, carbohydrates, saponins, and glycosides was recorded in the oils. These chemical compounds found in *C. nucifera* oil have several uses.

Neophytadiene is an anti-inflammatory agent; 2-pentadecanone 6,10,14-trimethyl is used in food flavorings and is a fragrance agent; hexadecanoic acid, methyl ester is used in cosmetics; 9,12-octadecadienoic acid methyl ester is also used as a flavoring agent; 9,12,15-octadecatrienoic acid, methyl ester is used in asthma management; 3,7,11,15-tetramethyl-2-hexadecene-1-ol is a medicinal agent; methyl stearate is used as an emulsifier and stabilizer; squalene reduces cholesterol level in animals and it is also suitable for oily skin dermatologically as it is lightweight

and not greasy; methyltris (tetramethylsilane) silane is a reagent used in organic synthesis; cyclotrisiloxane hexamethyl- is used as one of the ingredients in lotions; 1,1,1,3,5,5,5-heptamethyltrisiloxane is used in cosmetics line; and beta and alpha amyrin are anti-inflammatory, hepatoprotective and gastroprotective. The findings of this study are in line with a similar study by Ladokun *et al.* [17], who investigated the diabetic effect of phytocompounds synthesized from *Hunteria umbellata* using GC-MS analysis and molecular docking. Their study revealed 21 compounds, including 8,11-octadecadienoic acid and hexadecanoic acid, which were also encountered in this study. Abd'Rashed *et al*. [18] reported that the essential oils with a high percentage of monoterpenes exerted stronger antifungal activity.

Lauric acid, despite being the most abundant fatty acid in coconut oil, was not identified in this study. It could not be ascertained why this important component was not detected, as it wasn't the main focus of the study. However, this could be attributed to the sensitivity of lauric acid to heat and extended exposure during hot extraction, which could lead to partial degradation or reduced detectability [19, 20]. Fatty acids like lauric acid are not volatile enough for direct analysis via GC-MS. Detection can be limited without derivatization (e.g., conversion to methyl esters) [21]. Inadequate calibration of the GC-MS for short-chain fatty acids or overlapping peaks during analysis might also hinder detection. Ghani *et al*. [22] also pointed out that fatty acid composition may change depending on the extraction methods. Ajogun *et al*. [19] also buttressed this and found that the fatty acid profile was different in hot processed coconut oil.

The *C. nucifera* oil exerted vigorous activities against the test organisms as indicated by the varying zones of inhibition recorded during the study. The antimicrobial activities exerted by fluconazole, the hot-extracted and naturally-fermented VCO at 100 mg/mL against the test organisms showed no significant difference ($p = 0.07$). The ranges of the zones of inhibition at high concentrations (Table 3) are similar to that of Kannan *et al.* [23], who worked with *Candida albicans* and had average mean diameters of 19.6 mm and 23.5 mm. Khalid *et al*. [24] worked on the antifungal potential and antioxidant efficacy in the shell extract of *C. nucifera* against pathogenic dermal mycosis similarly. The extract was found to be effective against *Aspergillus niger* and *Aspergillus flavus,* which agrees with the findings of this study. Kumar [25] studied the influence of *Azadirachta indica*, *Melaleuca alternifolia*, and *C. nucifera* on *Candida albicans* strain in tissue conditioner at varying time intervals and reported that *C. nucifera* oil showed an inhibitory effect on the fungi which is similar to the findings of this study.

Udensi *et al.* [14] studied the antifungal properties of virgin coconut oil on *Candida albicans*, *Aspergillus niger*, and mold species, and it was noted that coconut oil was active against species of *Candida albicans* at a particular concentration. Shino *et al.* [26] isolated *Candida* species in children with ECC and studied the antifungal effect of coconut oil, probiotics, *Lactobacillus*, and 0.2% chlorhexidine on *C. albicans* compared to ketoconazole. The mean zone of inhibition for coconut oil was 16.8 mm, which shows that *C. nucifera* oil showed significant antifungal activity, just like the findings of this study.

5. Conclusion

This study demonstrated that virgin coconut oil (VCO), whether extracted with or without heat, exhibits antifungal activity and can effectively target pathogens such as *A. flavus*, *A. niger*, *C. albicans*, *C. krusei*, *Penicillium chrysogenum*, and *Trichophyton rubrum*. Its potential is particularly

significant in rising antimicrobial resistance, offering a natural alternative for managing fungal infections. The effective antifungal activities exerted by the *C. nucifera* oil could not be dissociated from the presence of the various active phytochemical compounds in it. The production of VCO could serve as a source of income for many people in tropical regions around the globe.

Future research could focus on optimizing the extraction methods of virgin coconut oil (VCO) to maximize its antifungal efficacy and the stability of its active compounds. Additionally, detailed mechanistic studies on how VCO inhibits fungal growth and its potential synergy with conventional antifungal agents could provide deeper insights, aiding its development as a complementary therapeutic option in the fight against resistant fungal infections.

Abbreviations

ANOVA	Analysis of variance
VCO	Virgin coconut oil
SPSS	Statistical package for the Social Sciences

Author Contributions

Authors OOB, ROA and TKB conceptualized the research. Authors OOB, ROA, TKB, MOF, MOO, and AO contributed to the development and writing of the manuscript. All authors contributed to validating, reviewing the manuscript, and approved the final version.

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Competing Interests

Authors declared no conflict of interest.

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