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# *Phytochemical analysis and Antibacterial activities of Coconut oil and water on Some Bacteria*

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**Abstract**

The phytochemical characteristics of coconut oil and water were examined, and the antimicrobial efficacy of coconut oil and water was assessed against several bacterial isolates. The discovered isolates included *Staphylococcus aureus, Escherichia coli, Bacillus sp,* and *Pseudomonas aeroginosa*. These isolates were identified by the examination of colony morphology, Gram staining, and biochemical assays. The test materials used in this study consist of extracted coconut oil and water, which were subjected to screening in order to identify and analyze their phytochemical composition. The phytochemical examination provided confirmation of the existence of alkaloids, saponins, tannins, and glycosides in both coconut oil and water. The sensitivity testing in this study used the agar disc diffusion assay, wherein the diameter of the zones of inhibition was evaluated in millimeters. Both coconut oil and water exhibited antibacterial properties against *Staphylococcus aureus*, *Escherichia coli*, and *Bacillus sp,* as shown by the formation of inhibition zones. The inhibition zones measured 10mm and 20mm for *S. aureus*, 9mm and 14mm for *E. coli*, and 12mm and 20mm for *Bacillus sp,* respectively. The bacterium *Pseudomonas aeruginosa* exhibited resistance to both coconut oil and water. Ciprofloxacin showed broad-spectrum antibacterial efficacy against all bacterial isolates included in this investigation.

**Key words: Phytochemicals, Antibacterial, Analysis, Coconut, Bacteria**

## INTRODUCTION

Plants of medicinal significance exhibit a wide range of phytochemicals that possess significant therapeutic qualities, making them valuable for addressing the challenges posed by developing illnesses. The investigation of medicinal plants used as cures in folklore has garnered significant interest within the scientific community as a means to address the issue of resistance to synthetic and conventional antimicrobials (Akinyele *et al.*, 2011). The development of antimicrobial resistance in microorganisms may be attributed to several factors, including the indiscriminate use of broad-spectrum antibiotics, immunosuppressive drugs, organ transplantation, and the usage of intravenous catheters (Selvamohan *et al*., 2012). In light of these issues, there is a collective endeavor to cultivate possibly efficacious, secure, and organic antimicrobial agents that may serve as alternatives to antibiotics for combating infections induced by organisms that have developed resistance to multiple drugs.

The plant species known as Coconut (*Cocos nucifera*) is classified under the botanical family Aracaceae. The plant is primarily used as a fundamental food crop

and serves as a valuable resource for wood and handicraft production, among many other applications. It is often regarded as the most versatile and medicinally significant plant in tropical and subtropical regions. Coconuts have been cultivated in tropical countries for over 4,500 years; nevertheless, they have seen a recent surge in popularity due to their distinctive taste, many culinary applications, and perceived health advantages. The coconut tree is often referred to as "the tree of life" due to its multifaceted use. It is a clear, colorless, sweet, and naturally flavored slightly acidic drink. Decades of research have shown that coconut water is a rich source of nutrient, including essential amino acids (lysine, leucine, cysteine, phenylalanine, tyrosine, histidine, and tryptophan), palmitic and oleic acids, and dietary minerals (Rukmini *et al.*, 2017). The coconut plant is grown due to its diverse range of applications, including both nutritional and therapeutic purposes. The coconut is an exceptional source of diverse natural compounds that have potential use in the production of pharmaceuticals and industrial goods. It has been shown to possess efficacy against a range of microorganisms, including fungus, bacteria, viruses, parasites, and dermatophytes (Florianna *et al.*, 2011). This research aimed to evaluate the phytochemical and bactericidal activity of coconut water and oil.

## MATERIALS AND METHODS

### Collection of Samples

The procurement of recently harvested, fully developed coconut fruits took place in the Eke Oko market, located in Anambra state. Subsequently, they were conveyed to the microbiology laboratory for the purpose of analysis.

### The process of extracting coconut oil

The working environment underwent a comprehensive cleaning, while the cutting instruments were meticulously sterilized. The coconut fruits underwent the process of being broken and dehusked. The extraction of the seed flesh from the shell was performed with a kitchen knife, followed by cutting and grading the flesh into smaller pieces. The flesh that had been assigned a grade was pulverized in a blender using warm water. Following the grinding process, the coconut milk was extracted from the chaff by carefully putting it into a sanitized plastic bowl via a sterile plastic sieve. The container of coconut milk was sealed and stored in a refrigerator at a temperature of 20°C for the duration of one night. The experiment included heating a mixture of coconut oil and water on a hot plate. The resulting mixture was then transferred to a sterilized stainless steel pot and stirred using a sterile plastic spatula. This process aimed to decrease the moisture content, resulting in the production of an aqueous extract of coconut oil. The agitation ceased after burnt fragments were apparent in the oil, prompting the pot to be placed aside for cooling until reaching a suitable temperature. The coconut oil underwent a filtration process using a sterile chiffon cloth in order to exclude any burnt particles. Subsequently, the filtered oil was carefully collected using a sterile plastic funnel and transferred into a sterile vial for further examination. The sample was kept at a temperature of 40°C in preparation for further analysis.

### The process of extracting coconut water

The coconut husk was fractured, allowing for the extraction of the coconut's fresh water, which was afterwards transferred into a container that had been sterilized.

### Phytochemical screening of the coconut oil and water

The present study focuses on the phytochemical screening of coconut oil and water.

#### i. Alkaloid Test

The experiment conducted was to test for the presence of alkaloids.

Approximately 3 mL of the extract was treated with a little quantity of Dragndorff's reagent. The formation of a red precipitate

serves as an indication for the presence of alkaloids.

**ii. Phenol Test**

The experiment conducted was aimed at testing the presence of phenols.

The extract was combined with a 2 ml aliquot of a 2% $FeCl_3$ solution. The presence of phenols may be inferred when seeing a coloring that is either blue-green or black.

**iii. Flavonoid Test**

The experiment included the combination of magnesium ribbon pieces with an extract, followed by the gradual addition of strong hydrochloric acid. The presence of flavonoids might be indicated by the colors orange, red, pink, or purple.

**iv. Glycoside Test**

In this experiment, a test was done to detect the presence of glycosides.

The extract was combined with 2 mL of glacial acetic acid solution containing 2 drops of 2% $FeCl_3$. The solution was transferred into a separate tube containing 2 milliliters of pure sulfuric acid. The presence of glycosides may be indicated by the observation of a brown ring during the interphase.

**v. Saponin Test**

The extract was subjected to boiling in the presence of 5 mL of distilled water for a duration of 5 minutes. The mixture underwent filtration while in a heated state. A volume of 1 milliliter (ml) of the filtrate was diluted by adding 4 ml of distilled water, followed by vigorous shaking. An observation was made about the stability of the broth while left undisturbed.

**Isolation of the bacteria**

The clinical samples were inoculated into plates containing Nutrient agar, Centrimide agar, Mannitol salt agar, Eosin methylene blue agar, and MacConkey agar using a sterile inoculation loop. The plates were subjected to incubation at a temperature of 37°C for a duration of 24 hours. Subsequently, the cultures were assessed for the presence of substantial growth. In order to create a pure culture, several colonies were subcultured.

**Identification of the bacteria**

Microscopic examination of bacteria that were isolated was conducted using the Gram stain method. The identification of bacterial isolates included a morphological analysis, whereby the colonies of these isolates were examined and characterized based on their forms, color, diameter, odor, and other distinguishing features (Macfaddin, 2000). Several biochemical tests were conducted to identify the bacteria, including the catalase test, indole test (Forbes *et al*., 2002), citrate test, coagulase test, oxidase test, and glucose test.

**Sterility Test**

The sterility test is a laboratory procedure used to assess the absence of viable microorganisms in a given sample.

The extracted coconut oil underwent culturing on MacConkey agar and Nutrient agar plates, followed by incubation at a temperature of 37°C for a duration of 24 hours. This process aimed to ascertain the purity of the oil and detect any potential contamination.

**Evaluation of Antibacterial Activity**

The preparation of bacterial inocula included the transfer of bacterial colonies using a sterile inoculating loop, followed by their suspension in a 10ml volume of nutritional broth. The inocula were adjusted to a 0.5 MacFarland standard and were put onto a gelled agar plate using swab sticks. Subsequently, the plate was left undisturbed. Discs of filter paper measuring 6mm in diameter were immersed in the necessary concentration of test samples and afterwards positioned onto the agar plates. The plates were subjected to incubation at a temperature of 37°C for a duration of 24 hours. During this period, the plates were carefully examined to identify any zones of inhibition, measured in millimeters, that were formed as a result of the application of coconut oil and water. The use of

ciprofloxacin was employed as a control for comparison.

**RESULTS**

The phytochemical content identified in the examined samples of coconut oil and water is reported in Table 1. Table 2 displays the cultural features and biochemical test results of the isolates. The oil that was obtained was cultivated on Nutrient agar and MacConkey agar, and the absence of growth seen indicates good sterility outcomes. Table 3 displays the results of the sterility test conducted on coconut oil.The present study aimed to assess the antibacterial efficacy of coconut oil and water against four bacterial strains, including *Staphylococcus aureus*, *Escherichia coli*, *Bacillus sp*, and *Pseudomonas aeruginosa*. Ciprofloxacin was used as the control in this investigation. Table 4 presents the observed zone of inhibition resulting from the application of coconut oil and water on the test organisms.

**Table 1: Phytochemical analysis of coconut oil and water**

| Phytochemicals | Coconut oil | Coconut water |
|---|---|---|
| Alkaloids | + | +++ |
| Flavonoids | - | - |
| Glycosides | ++ | ++ |
| Saponins | +++ | ++ |
| Tanins | + | +++ |

**Keys:** +++: Highly present, ++: Moderately present, +: Slightly present, -: Absent.

**Table 2: Biochemical reactions of the isolates**

| Cultural characteristics | Morphology | Gram stain | Catalase | Citrase | Coagulase | Oxidase | Indole | Motility | Glucose | Lactose | Sucrose | Probable organism |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Greenish metallic sheen on EMB agar | Rod | - | + | - | - | - | + | + | + | + | + | *Escherichia coli* |
| Yellow colonies on Mannitol agar | Cocci | + | + | + | + | - | - | - | + | + | + | *Staphylococcus aureus* |
| Very large whitish colonies on Nutrient agar | Rod | + | + | + | + | - | - | + | + | - | - | *Bacillus sp* |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Greenish blue colonies on Centrimide agar | Rod | - | + | + | - | + | - | + | - | - | - | *Pseudomonas aeruginosa* |

**Keys**: +: Positive, -: Negative.

**Table 3: Sterility test of coconut oil sample**

| Media | Temperature of incubation | Number of colonies | Inference |
|---|---|---|---|
| Nutrient agar | $37^0$C | 0 | Satisfactory |
| MacConkey agar | $37^0$C | 0 | Satisfactory |

**Table 4: Antibacterial activity of coconut oil and water on isolates**

| Bacteria | Coconut water | Coconut oil | Ciprofloxacin(control) |
|---|---|---|---|
| *Escherichia coli* | 14mm | 9mm | 30mm |
| *Staphylococcus aureus* | 20mm | 10mm | 30mm |
| *Bacillus sp* | 20mm | 12mm | 35mm |
| *Pseudomonas aeruginosa* | Nil | Nil | 10mm |

## DISCUSSION

Phytochemicals have been discovered to have a diverse array of properties that might potentially contribute to the prevention of chronic illnesses. The phytochemical analysis of coconut oil and water revealed the presence of alkaloids, saponins, tannins, and glycosides, whereas the absence of flavonoids was observed. The findings shown in Table 1 are consistent with the findings reported by Obidoa *et al.* (2010), indicating the moderate presence of tannins, alkaloids, saponins, and glycosides in coconut oil and also which indicates the presence of alkaloids, tannins, saponins, and glycosides in coconut water.

The purpose of this study was to examine the antibacterial properties of coconut oil and water against several bacterial isolates, including *Staphylococcus aureus*, *Escherichia coli*, *Bacillus spp*, and *Pseudomonas aeruginosa*. The mentioned isolates have been documented as prevalent pathogens that are responsible for urinary tract infections (Odoki *et al*., 2019) and nosocomial infections (Khan *et al*., 2019), among other conditions. It is worth noting that *Bacillus sp*. is recognized as an opportunistic pathogen in individuals receiving hospital care. The efficacy of coconut oil and water in inhibiting bacterial growth was shown in all test organisms, with the exception of one. This research demonstrates that *Bacillus spp* has the greatest vulnerability to coconut oil, whilst *Escherichia coli* displays the lowest susceptibility, and *Pseudomonas aeroginosa* exhibits resistance. The findings shown in Table 4 align with the prior study conducted by Effiong *et al*. (2018), which reported notable antibacterial effects of coconut oil against *Staphylococcus aureus* and *Escherichia*

*coli*. The findings of Florianna *et al*. (2015) about the efficacy of coconut oil against *Escherichia coli, Staphylococcus aureus*, and *Bacillus* sp are consistent with the results obtained in this research.

The bacterial isolates, including *Staphylococcus aureus, Escherichia coli*, and *Bacillus sp*, shown no resistance to coconut water. The potential correlation might be attributed to the existence of phytochemicals inside coconut water. The research conducted by Fowoyo and Alamu (2018) supports these results, as they observed the antimicrobial effects of coconut water against *Staphylococcus aureus, Escherichia coli*, and *Bacillus sp*. Nevertheless, the findings presented in this research contradict the conclusions drawn by Rukmini *et al*. (2017), which suggested that coconut water does not possess antibacterial properties against microbes.

Table 4 presents the results of the antibacterial activity shown by ciprofloxacin (control). The inhibition zone diameters for several bacterial strains were recorded as follows: 30mm for *E.coli*, 30mm for *S.aureus*, 35mm for *Bacillus sp*, and 10mm for *P.aeroginosa*.

In conclusion, it can be inferred that the aforementioned points collectively support the notion that.The motivation for the initiation of this investigation stemmed from the escalating phenomenon of antibiotic resistance. Plant extracts have recently gained attention as potential antibacterial agents due to their rich assortment of phytochemicals with significant therapeutic capabilities that might be used in the management of both developing and re-emerging illnesses.

This research presents an analysis of the phytochemical composition of coconut oil and water, highlighting their biological activities and confirming their pharmacological qualities. This study also demonstrates the antibacterial capabilities of coconut oil and water, highlighting their potential use in the formulation of antibacterial soaps and lotions.

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