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Original Research

Antimicrobial Activity of Coconut Oil and its Derivative (Lauric Acid) on Some Selected Clinical Isolates

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Abstract: This study investigates the in vitro antimicrobial activity of coconut oil and its fatty acid(lauric acid) on selected clinical isolates. Clinical isolates were obtained from the General Hospital Maitama, Abuja, Nigeria. Media preparation and biochemical examination of the organisms were done according to standard methods. Organisms used were StaphylococcusaureusStreptococcus species, Lactobacillus species and Escherichia coli. Coconut oil was extracted through fermentation method were as lauric acids was esterified from coconut oil through freezing and were subjected to sterility test. Bauer-Kirby disc diffusion assay was used for the sensitivity assessment. Zones of inhibition were measured in diametre. Coconut oil showed resistant on the isolates at the various dilution concentrations. Lauric acid demonstrated significantly appreciable antim icrobial effect on the test organisms with the highest zone of inhibition on Staphylococcus aureus (10.50)mm, Streptococcus speci es (10.00) mm, Lactobacillus species (10.00) mm and the lowest inhibition on Escherichia coli (4.00)mm even at the Minimum Inhibitory Concentration (MIC).Escherichia coli which showed relatively low zone of inhibition even at the highest dilution concentration. The acid generally demonstrated appreciable sensitivity on the isolates with low effect on E. coli compare to other strains. This study recommends the use of coconut oil as therapeutic agent as well as in fighting antibiotic resistant since it contains lauric acid which is bactericidal. Further studies should be done on the oil and its derivative both in vitro and in vivo unveils its mechanisms of actions.

Keywords: Antimicrobial activity, coconut oil, lauric acid, clinical isolates.

INTRODUCTION

Plants of medicinal importance containhuge varieties of phytochemicals with important therapeutic properties that can be used in the treatment of emerging and re-emerging diseases. Consequently, there is the increasingly justified assumption which claims that traditional medicine is cheaper and more effective than modern medicine. The studies of medicinal plants used as folklore remedies have therefore attracted immense attention in the scientific world in an attempt to find possible solutions to the problems of multiple resistances to the existing synthetic and conventional antimicrobials (Taiwo*et' al.,* 2011). The discovery of antibiotics had eradicated the infections that once ravaged the humankind, but their indiscriminate use has led to the development of multidrug-resistant pathogens (Shanmugan*et' al.,* 2008).

Coconuts are an underutilized food with a hidden wealth of nutritional value for the body. The fat content plays into the mass confusion surrounding healthy and unhealthy fats, but there are a surprising number of benefits with this unusual nut as it provides a very unique type of oil, made of several ingredients including medium chain fatty acids, lauric acid and saturated fat (Schlievert,*et' al.,* 2008). It is semi-solid at room temperature as a soft, almost waxy substance. Coconut oil is prized for its health-giving properties, considered one of the beneficial oils to use when cooking. Coconut oil is stable in high heat while many other oils are damaged upon heating, making them very unhealthy for cooking. Over the past several years, nutritional advice has focused on the avoidance of fat, particularly saturated fat. We are now learning, or relearning, what many cultures have known for centuries. Healthy fats can include some saturated fat. The quality of animal fats will depend on the health of the animal. We are also learning that many vegetable oils that were once considered healthy are known to become damaged with heat. One of the amazing qualities of coconut oil is its antibacterial properties. Monolaurin, an ingredient in coconut oil, has long been recognized for its bug-fighting properties. It is found in breast milk, perhaps in part to help protect the developing baby from infection (Clarke and May, 2007). It appears that coconut milk can protect against several different kinds of bacteria and fungi of clinical impact and can further benefit the skin by treating and preventing skin infections (Carpo et' al., 2007; Clarke and May, 2007). According to Abbas *et' al.* (2017), this virgin coconut oil which is a potent nondrug or natural yeast fighter, contains three medium chain fatty acids, *i.e*., lauric acid (50–53%), caprylic acid, and capric acid, all of which have antibacterial and antifungal effect against lipid coated bacteria such as staphylococcus species and fungi such as *Candida spp*.

Lauric acid a twelve (12) carbon chain acids, is one of the medium chain fatty acids gotten from some plants oil particularly coconut oil and others related oil such as palm kernel oil which has been known as one of the most active ingredient and is more predominant in the total saturated fat present (Bruce, 2000).This acid is found in many vegetables, fats particularly in coconut oil and palm kernel oil (Chuah e*t' al.*, 2014); and has been known as one of the most active ingredient and composed over 52% of the total 92% saturated fats present in the coconut oil and is claimed to play a significant role in the healing miracle that is revealed in coconut oil (Fife, 2003). Medium-chain free fatty acids which lauric acid fall under have been found to have a broad spectrum of microbicidal activity though the mechanisms by which the lipids kill bacteria is not known, but electron microscope studies indicate that they disrupt cell membranes(Ogbolu, 2007). On the other hand, Free Fatty Acids (FFA) of various chain lengths (C_8- C_{18}) have antibacterial activity against a range of Gram-positive bacteria, but not against a number of Gram-negative bacteria Georgel *et al.*, 2005;Skrivanova *et al.*, 2005;Drake *et al.*, 2008). Variations in composition of plant and genetic disparity among bacteria and fungi of the same or different species have been found to be responsible for the few inconsistencies in the antibacterial and antifungal properties of plant extract. The esterification of coconut oil which yielded a carbon chain has proved beyond reasonable doubt that, lauric acid 12-carbon chain fatty acid is more biological active and has the highest antiviral activities than coconut oil which is the parent substance (Kabara, 1960). This resulted from the Medium Chain Triglycerides (MCTs) present in coconut oil which anti-bacterial influence because it has the ability to disintegrate bacterial cell walls; MCTs are also presenting the ability to treat severe bacterial infections that are antibiotic resistant (Bruce, 2000). Despite the vast impact of coconut plants as a whole and its health importance to humanity hitherto, most people still lack the basic knowledge in this plants and relatively few studies has been done to ascertain its health impact.In this study, antimicrobial activity of coconut oil and its derivative (lauric acid) were investigated.

TERIALS AND METHODS

Preparation of coconut oil

Fresh coconut (*Cocos nucifera*) was obtained from Lafia modern market Lafia, Nigeria. The fresh coconut meat was grated and pressed using a sterilized sieve to produce coconut milk, which was further allowed to ferment for 48 hours under anaerobic condition (Abbas *et' al.,* 2017). After the fermentation, three layers were formed: the water layer, lipid layer and the protein coat layer. Protein coat and the water layer were separated from the oil (lipid layer). The oil was then heated slightly to remove remaining moisture. After which the oil was filtered by passage through a 25m-pore size filter (Millipore, St. Quentin, France) to give an aqueous extract of coconut oil. This was collected in a sterile vial and stored at 4°C until use.

Preparation of lauric acid

Extra virgin coconut oil was poured into a temperature glass container, manufacturer filter to remove impurities, digital freezer was set at 25.1^{o}C 3 to freeze coconut oil and lauric acid was extracted at 47^{o} C (Abbas *et' al.,* 2017).

Suspension of test organisms

Suspension of each of the test organisms was made by collecting a loopful of colony from each plate and was incubated overnight at 37°C in Nutrient broth. The overnight broth culture of organisms was diluted in nutrient broth to an inoculum load of approximately $1x10^6$ cfu/ml. It was standardized according to National Committee for Clinical Laboratory Standards (NCCLS, 2002) by gradually adding normal saline to compare its turbidity to McFarland turbidity standard of 0.5 which is approximately 1.0×10^6 cfu/ml. Sterile swab sticks were dipped into each of the bacterial solution and were used to inoculate the solidified Nutrient agar plates ensuring that the plates were completely covered for uniform growth as described by (Aboh *et al*., 2013).

Sterility test

Pure virgin coconut oil and the extracted lauric acid were cultured differently on prepared media plates and incubated overnight at 4°C. This was done to ensure that the extracts were completely sterile. All media prepared were picked at random and incubated overnight at 37°C for the purpose of the study.

Antimicrobial susceptibility test

Antimicrobial susceptibility test was carried out in each of the plate using agar disc diffusion method as described by Bauer-Kirby (2008).This involves a heavy inoculation of an agar plate with the test organisms. A disc of filter paper (Whatman filter paper) was impregnated with a known volume and appropriate concentration of lauric acid and was placed on a plate of susceptibility testing agar uniformly inoculated with the test organism and equally spaced on the inoculated plate. The antimicrobial agent diffused from the disc into the medium and the growth of the test organism was inhibited at a distance from the disc that is related (among other factors) to the susceptibility of the organisms. Strains susceptible to the antimicrobial were inhibited at a distance from the disc whereas resistant strains have smaller zones of inhibition or grow up to edge of the disc (Cheesbrough, 2006). Following incubation, the agar plate was examined for zones of inhibition (areas of no growth) surrounding the discs. Zone of inhibition indicates antimicrobial activity against the organisms. Absence of zone of inhibition indicates that the acid was ineffective against the test organisms or the organisms are resistant to the acid.

RESULT AND DISCUSSION

Result of the morphological identification, biochemical reaction, carbohydrate utilization and haemolytic reaction of the test organisms is shown in table 1 below.

Table 1:**Biochemical identification and carbohydrate utilization of the isolates**

Biochemical examination	***Staphylococcus ureus***	***Streptococcus species***	***Lactobacillus species***	***Escherichia coli***
Catalase	+	-	-	+
Oxidase	-	-	-	-
Coagulase	+	-	-	-
Motility	-	-	-	+
Indole	-	-	-	+
Methyl Red	+	+	-	+
V. P	+	+	-	-
Urease	+	-	-	-
Glucose	+	+	-	+
Sucrose	+	+	-	+
Mannose	+	+	-	-
Mannitol	+	-	-	+
Lactose	+	+	+	-
Haemolysis	+	+	-	-
Acid fast stain	-	-	+	-

Keys: + = Positive; **- =** Negative; VP = Voges Proskauer

The result of the agar disc diffusion antimicrobial assay of coconut oil on the selected clinical isolates is shown in table 2 below. The clinical isolates used for the sensitivity assay were*:* *Staphylococcus aureus, Streptococcus species,Escherichia coli*and *Lactobacillus*species showing resistance to the oil extract.

Table 2: **Sensitivity assay of coconut oil on the isolates**

BACTERIAL ISOLATES	**30%**	**50%**	**70%**	**100%**
S. aureus	R	R	R	R
Streptococcus	R	R	R	R
E. coli	R	R	R	R
Lactobacillus	R	R	R	R

R = Resistance of isolates to the oil extract, % = percentage of the dilution concentration

The result of the agar disc diffusion antimicrobial assay of coconut oil on the selected clinical isolates is shown in table 2 below. The clinical isolates used for the sensitivity assay were*:* *Staphylococcus aureus, Streptococcus, Escherichia coli*and *Lactobacillus* the zones of inhibition observed were recorded accordingly.

Table 3: **Sensitivity assay of coconut oil on the isolates**

BACTERIAL STRAINS	**Susceptibility at giving concentration**			
	100%	**70%**	**50%**	**30%**
Staphylococcus aureus	10.50	9.00	8.50	7.50
Streptococci	10.00	9.00	8.50	7.00
Escherichia coli	4.00	3.00	2.00	1.50
Lactobacilli	10.00	9.50	10.00	8.00

% = Percentage

Table 4 below is the presentation of the result of Minimum Inhibitory Concentration (MIC) of lauric acid sensitivity on the isolates at various dilution concentrations.

BACTERIAL STRAINS	**Minimum Bactericidal Concentration (MBC) (30% concentration)**			
Staphylococcus aureus	4.00	3.05	2.50	2.00
Streptococci	4.00	3.00	2.50	2.00
Escherichia coli	1.50	1.0	0.20	0.05
Lactobacilli	4.00	3.00	2.05	2.00

Table 4: **Minimum Inhibitory Concentration (MIC)**

In this study, both coconut oil and its fatty acid (Lauric acid) were for their antimicrobial properties on a few selected clinical isolates. Organisms isolated were *Staphylococcus aureus, Streptococcus, Escherichia coli*and *Lactobacillus species*. Both organisms showed resistance to coconut oil at the variousdilutionconcentrations as opposed to the study of Ogbolu *et' al.*(2007), who reported the antimicrobial potential of coconut oil on fungal organisms. The method employed by Ogbolu *et' al.* (2007) differed from this study because coconut oil was diluted with 1% ethanol which earlier knowledge has educated us on the antimicrobial properties of all classes of alcohol in which ethanol is included. The diluent could be responsible for the inhibitory effect exerted in the study.

Lauric acid in this study showed considerable inhibitory effect on virtually all the clinical isolates used in this studywith decrease in effects corresponding to the concentration of the acid. The acid demonstrated highest zones of inhibition on the isolates with the following diametre: *Staphylococcus aureus* (10.50)mm, *Streptococcus species* (10.00)mm *Lactobacillus* *species* (10.00)mm whereas the lowest inhibitory effect was observed on *Escherichia coli*(4.00)mm at the same dilution concentration. In general the acid was more effective against *Staphylococcus aureus,Streptococci, and Lactobacilli*even at the lowestdilution concentration whereas *E. coli* which is a Gram negative bacterium showed relatively low inhibition.Similarly Abbas *et' al.* (2016), reported that Synthetic sodium laurate (lauric acid) fatty acid exhibit significantly high antimicrobial activity by inhibiting microbial survival and biofilm growth against *Streptococcus mutans*. Arguably, Padgett *et' al.* (2000), reported that high level of lauric acid addition (8%) significantly lower the film water permeability. This result conforms to the popular assertion that says the higher the concentration, the higher the antimicrobial effect of agent against organisms (Rutala *et al.*, 2008). *Escherichia coli*which is Gram negative bacteria showed very low inhibitory effect to the acid tested at a lower concentration compare to other Gram positive bacteria such as *S. aureus*, *Streptococci* and *Lactobacilli*. This finding obeysthe findings of Mamman *et al.* (2005) that says Gram negative bacteria exhibit much resistance compare to Gram positive bacteria. Lauric acid exhibited appreciably high antimicrobial activity in some clinical isolates than others and the zones of inhibition varied based on their dilution concentration declining as the dilution concentration decreases.

CONCLUSION AND RECOMMENDATION

This study argued the earlier acclamation that says coconut oil has antimicrobial activity in vitro and further affirmedthat, lauric acid has antibacterial effect on Gram positive bacteria more compare to Gram negative bacteria. This however recommends use of lauric acid in treating some of the emerging and re-emerging diseases as well as improving health status. More studies should be done to ascertain the mechanisms of actions of this acid on microorganism generally and their susceptibility pattern.

Declarations

Authors' contributions

AAA is the main author and was responsible for the writing of the manuscript, participated in data collection and interpretation as well as drafting and review of the manuscript. UP was involved in the study design and data interpretation. EBEA and TTK reviewed the manuscript. Both authors read and approved the final manuscript.

Ethical consideration

This research does not required ethical clearance as human participants were not involved in the study; however, proper permission was taken to obtain clinical isolates for the purpose of the study.

Competing interests

There is no competing interest in the publication of the journal by authors.

Availability of data and materials: The datasets used and analyzed during this study are available from the corresponding author on reasonable request.

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REFERENCES

Abbas, A. A.; Peter, U.; Akeh, M.; Adeola, J.; Ewenighi C. O. and Ishaku, A. (2017). Antibacterial Activity of Lauric Acid on Some Selected Clinical Isolates. *Annals of Clinical and Laboratory Research.*(10). 2386-5180.

Abdulelah H. Al-Adhroey, Zurainee M. Nor, Hesham M. Al-Mekhlafi, Adel A. Amran, Rohela Mahmud. (2011). Evaluation ofthe use of *Cocosnucifera* as antimalarial remedy in Malaysian folk medicine.*Journal of Ethnopharmacology.*134:3, 988-991.

Aboh, M. I., Oladosu, P. and Ibrahim, K. (2013). Antimicrobial Activities of Some Brands of Household Disinfectants Marketed In Abuja Municipal Area Council, Federal Capital Territory, Nigeria. *American Journal of Research Communication*, 1(8): 172-183.

"Define Abuja's at Dictionary.com".dictionary.com. Random House, Inc. *Retrieved 2015.*

Bruce, F. (2000). Coconut oil and Medium Chain Triglyceride. Health wise Chapter 7.

Carpo, BG., Verallo-Rowell, VM., Kabara, J. (2007). Novel antibacterial activity of monolaurin compared with conventional antibiotics against organisms from skin infections: an in vitro study. J Drugs Dermatol, 6(10):991-8.

Cheesbrough, M. (2006). *District Laboratory Practice in Tropical Countries Part 1*, (2nd Ed). New York:Cambridge University Press. Pp: 38-39.

Chuah, G., Rozanna, T. D., Salmiah, A., Thomas, Y. C. and Sa'ari, S. M. (2014). Fatty acids used as Phase change Materials (PCMs) for Thermal Energy storage in building Material Application. University publication Malaysia.

Clarke, NM., May, JT. Effect of antimicrobial factors in human milk on rhinoviruses and milk-borne cytomegalovirus in vitro.

Drake, D. R.; Brogden, K. A.; Dawson, D. V. and Wertz, P. W. (2008) Thematic review series: skin lipids. Antimicrobial lipids at the skin surface. *Journal of Lipid Residue*.49:4–11.

Fife, B. (2003). *The Healing Miracles of Coconut Oil*,Avery Publishing Group Inc., New York.

Georgel P, Crozat K, Lauth X, Makrantonaki E, Seltmann H, Sovath S (2005) A toll-like receptor 2-responsive lipid effector pathway protects mammals against skin infections with gram-positive bacteria. *Journal of Infections and Immunology*.73:4512–21.

Ja-Hyung L.and Young-Wook, J. (2016).Antimicrobial Effect of a Lauric Acid on Streptococcus MutansBiofilm.*Annals of International medical and* DentalResearch.2(4):21.

Kabara, J. J.Swieczkowski, D. M.; Conley, A. J. and Truant, J. P. (1972)Fatty acids and Derivatives as Antimicrobial Agents:*Antimicrobial Agents Chemotherapy*. 2:23–28.

Kabara, J. J.(1978). Fatty Acids Derivatives as Antimicrobial Agents-A Review-Symposium on Pharmacological Effects of Lipids. AOCSP113.

Kitahara T, Koyama N, Matsuda J, Aoyama Y, Hirakata Y, Kamihira S(2004)Antimicrobial activity of saturated fatty acids and fatty amine s against methicillin resi stant *Staphylococcusaureus. Biological and Pharmaceutical Bulletin*.27:1321–1326.

Mamman, P.H., Kazeem, H.M. and Kwanashie, C.N. (2005). Disinfectant effect of carcil(Alkyl- benxyldimethyl ammonium chloride) on bacteria. *Sciences,* 1:33 – 136.

Manisha DebMandal, Shyamapada Mandal. (2011). Coconut (*Cocosnucifera arecaceae*): In health promotion and diseaseprevention. *Asian Pacific Journal of Tropical Medicine* 4:3, 241-247.

Murray, S. (2011) "Life of poverty in Abuja's wealth." *BBC.* Tuesday 13 February 2007. Retrieved on 12 September.

National Committee for Clinical Laboratory Standard. (2002). Performance standard for antimicrobial disc susceptibility testing. Twelfth International Supplement.

Ogbolu, D. O., Oni, A. A., Daini, O. A., and Oloko, A. P. (2007). *In Vitro* Antimicrobial Properties of Coconut Oil on *Candida* Species. *Journal of Medicinal Food.*(10)2:384–387.

Padgett, T.; Han, L.and Dawsow, P. L. (2000). Effect Of Lauric Acid Addition On The Antimicrobial Efficacy And Water Permeability Of Corn Zein Films Containing Nisin *Journal of Food Processing and Preservation.*29634-0371

Rutala, W.A. and Weber, D.J. (2013). Disinfection and Sterilization. *American Journal of Infection Control.* 41(5):S2 –S3.

Seidele, A. and Linke, W. F. (2014). *Solubilities of Inorganic and Organic Compound,* New York. D. Van Nostrand Company. PP 742-743.

Shanmugan, H.; Anil, K.; Mukesh, D. Synergism between natural products and antibiotics against infectious diseases. J. Phytomed. 2008, 15, 639-652.

Schlievert, PM., Strandberg, KL., Brosnahan, AJ., et al. (2008). Glycerol monolaurate does not alter rhesus macaque (Macaca mulatta) vaginal lactobacilli and is safe for chronic use. Antimicrob Agents Chemother, 52(12):4448-54.

Skrivanova, E.; Marounek, M.;Dlouha, G. andKanka, J. (2005) Susceptibility of Clostridium perfringens to C-C fatty acids. *Letter of Applied Microbiology*. 41:77–81.

Taiwo, A. A.; Omobola, O. O; David, A. A. and Anthony, I. O. (2011) In-Vitro Antibacterial Properties of Crude Aqueous and n-Hexane Extracts of the Husk of *Cocos nucifera: Molecules.* (16): 2135-2145.

Teruaki,N.; Mandy, C. K. and Chun-Ming, H. (2009). Antimicrobial Property of Lauric Acid, Against *Propionibacterium acnes*: It’s Therapeutic Potential for Inflammatory Acne Vulgaris: *Journal of Investigative Dermatology*. 129.

Wille, J. J. and Kydonieus, A. (2003) Palmitoleic acid isomer (C16:1delta6) in Humanskin Sebum is Effective against Gram-positive bacteria.Skin PharmacolAppl Skin Physiol16:176–188.

Yoo, Songkro, AnusakSirikatitham, SupreedeeSungkarak, Khemmarat Buaking, Juraithip Wungsintaweekul, DuangkhaeManeenuan, KwunchitOungbho (2010).Characterizatio of Aromat herapy Massage Oils Prepared from Virgin Coconut Oiland SomeEssential Oils. *Journal of the American Oil Chemists' Society* 87:1, 93- 107.