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ANTIMICROBIAL PROPERTIES OF *Cympobogon citratus* AND *Cocos nucifera* OILS

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ABSTRACT

Cympobogon citratus and *Cocos nucifera* oil were evaluated for their antibacterial and antifungal activity against pathogenic strains of *Klebsiella pneumoniae* ,*Salmonella typhi*, *Esherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* ATT25667 and *Candida albicans*(the only fungi strain).The antimicrobial properties against the aforementioned strains were compared to standard antibiotics, third generation fluoroquinolone ciprofloxacin and imidazole respectively. The *Salmonella typhi* strain showed higher susceptibility to *Cympobogon citratus* oil while *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* showed appreciable susceptibility. In *Cocos nucifera* oil, the *Candida albicans* showed a greater susceptibility when compared to the standard imidazole.The *Cocos nucifera* oil showed greater susceptibility to *Staphylococcus aureus* and *Eshcerichia coli* strain. Therefore the oil extract of these *Cympobogon citratus* and *Cocos nucifera* have been found to have potential application for prevention of bacteria and fungi infection. The present study supports its traditional uses.
Keywords: antibacterial, antifungal, antiseptic, antibiotic

INTRODUCTION

Traditional medicines and herbal plants had been in existence long unrecorded years ago. The practice and the use of traditional medicines and herbs in Nigeria is now coming to the fore since various health workers are coming to realize the benefits of natural herbs to synthetic drugs. The problems that have been facing traditional medicine since inception is the dosage of the herbs recommended for various treatments of the diseased conditions, the hygienic level of such herbs during preparation and the side-effects that long-term use of the herbs pose to the health of individuals. Among the diseases that have been successfully managed traditionally are malaria, epilepsy, convulsion, typhoid fever, fungal infections and worm infections. (Sofowora, 1996).

Industrial interest in exploiting plants for medicinal purpose is exclusively found in China and Japan (Cheji,1988). Cymbopogon citratus has been found to be antiseptic, fungicidal, diuretic, and externally against athlete foot and acne (Sofowoa, 1996).Cymbopogon citratus oil has been found to consists mainly of citral.citral is a mixture of two stereo isomeric monterpene aldehydes (Onawunmi et al, 2007). Cocos nucifera oil is another therapeutic oil that has been found to be effective against peptic ulcers, genital herpes and skin infection.(Sofowora,1996).it has been found to be antifungal, antiviral and antibacterial(Sofowora,1996).

This study was undertaken to determine the antibacterial and antifungal activities of Cymbopogon citratus oil and Cocos nucifera oil. The research is aimed at enhancing available knowledge on the antimicrobial .The research is aimed at enhancing available

knowledge on the antimicrobial analysis of the oil with a view to promoting these natural herbs and to optimize the use of available natural resources in the environment

MATERIALS AND METHODS

The Cympobogon citratus leaves and the Cocos nucifera fruit were obtained from a local market (Itoku) in Abeokuta, Ogun state.

Micro-Organisms

Pure specimens of *Klebsiella pneumoniae*, *Staphylococcus aureus* ATT25667, *Salmonella typhi*, *Pseudomonas aeruginosa*, *Esherichia coli*, and *Candida albicans* (10^{-6}cfu/ml) were obtained from microbiology department of sacred heart hospital.Abeokuta.

Extraction of Oil

The oil from the *Cympobogon citratus* and *Cocos nucifera* was extracted by steam extraction methods.100ml of sterile distilled water was dispensed into a clean conical flask containing 10g of powdered sample and stirred every 30minutes for 3hrs.and allowed to stand for 24hrs before extraction. The two mixtures were filtered using whatman's filter paper no 1 and the filtrate was concentrated under a reduced pressure in a rotary evaporator until a semi-solid remains.

Minimum Inhibitory Concentration

After extraction, the extract was diluted in the nutrient broth to obtain different dilution of extract. A drop each of microbial suspension in a pure state was added to each dilution and incubated at 37ºc for 18-24hrs. The mic dilution was determined by incorporating constant volumes 0.2ml of each dilution of the extract into the punch-holes on a pre-seeded appropriate agar medium. aliquot of the extract 0.2gm was dissolved in 100ml of sterile distilled water to obtain 2,0mg/ml. the concentration was then doubly diluted in sterile distilled water to obtain concentrations of ½,1/4,1/8,1/16,1/32,and1/64mg/ml . (Tuhin, et al, 2007)

The growth of the test organism in each dilution of cympobogon citratus oil and cocos nucifera oil was examined and compared with controls by matching their turbidity .the clear preparation was considered as no growth. The lowest concentration at which bacteria and fungi were inhibited as judged by lack of turbidity was considered as minimum inhibitory concentration (mic) and was recorded.

Minimum Bactericidal and Fungicidal Concentration

Following the incubation and reading of mic results, the adhesive seal was removed and the tubes showing no visible growth were subcultured on blood agar plates. The plates were incubated overnight at 37ºc and examined for growth. The minimal bactericidal and fungicidal concentrations were read as the lowest concentration which yields no growth upon subculture.

Antimicrobial Susceptibility using Agar Diffusion Method

A dilution of extract was prepared at their different mic and poured to the disc (made from whatman no1 filter paper).the disc containing mic extract and third generation fluoroquinolone ciprofloxacin (5µg/disc) and imidazole(20µg/disc).were placed on the pre inoculated plate with test organisms with a sterile forceps and each of them was slightly

pressed against agar surface and then incubated aerobically at 37ºc for 24hours.the diameter of zone of inhibition denoted the relative susceptibility to a particular antimicrobial agent which was detected by a formation of a clear zone around the disc .the diameter of inhibition was measured in millimeter under the surface of the petridish using a transparent scale

RESULTS

The results reveals that the minimum inhibitory concentration value of *Salmonella typhi* was the lowest 3.9µg/ml, followed by that of *Klebsiella pneumonia* and *Staphylococcus aureus* ATT25667 (7.8) respectively.*Escherichia coli* followed suit with minimum inhibitory concentration of 15.6µg while *Pseudomonas aeruginosa* was the least susceptible to the *Cympobogon citratus* extract with minimum inhibitory concentration of 31.3µng/ml in Table 1.

The antimicrobial susceptibility profile of *Salmonella typhi* in table 2 was found higher in *Cymbopogon citratus* 15mm than that of standard ciprofloxacin 14mm.also in table 2, *Klebsiella pneumonia* also showed a higher susceptibility to *Cympobogon citratus* with 15mm compared with 20mm of the standard drug. The fungi, Candida albicans showed high zones of inhibition 30mm compared to the standard imidazole 14mm.

In *Cocos nucifera, Escherichia coli* has the highest minimum inhibitory concentration value of 31.3µg/ml in table 3 while *Klebsiella pneumoniae* and *Staphylococcus aureus* had the lowest minimum inhibitory concentration value of 7.8µg/ml respectively. *Salmonella typhi* and *Candida albicans* had minimum inhibitory concentration value of 15.6 respectively.

In Table 4, *Cocos nucifera* oil showed a greater zone of inhibition for *Staphylococcus aureus* 33mmto the standard ciprofloxacin of 30mm .*Pseudomonas aureginosa* had a zone of inhibition of 32mm to ciprofloxacin of 30mm.*Cocos nucifera* oil showed an impressive zone of inhibition of 20mm against the standard imidazole 14mm for *Candida albicans*.
Tables 5 and 6 showed the minimal bactericidal concentration and minimal fungicidal concentration for *Cympobogon citratus* and *Cocos nucifera* respectively. This showed the lowest concentration of extract that was active against the microorganisms used for the research.

Table 1: Minimum Inhibitory Concentration (Mic) Of *Cympobogon Citratus* Oil Against Different Organisms.

Micro-organisms	MIC dilution ratio	MIC value (µg/ml)
Klebsiella pneumonia	1:32	7.8
Staphylococcus aureus	1:32	7.8
Pseudomonas aeruginosa	1:8	31.3
Escherichia coli	1:16	15.6
Salmonella typhi	1:64	3.9
Candida albicans		62.5

Note: Values are average of three observations

TABLE 2: Diameter of Zones of Inhibition for Cympobogon Citratus against Test Organisms

Extract/disc	k. pneu		s. aureus		p. aerugi		E. coli		S. typhi		C .albicans	
	D	Z	D	Z	D	Z	D	Z	D	Z	D	Z
Cympobogon Citrates	7.8	15	7.8	17	31.3	20	15.6	17.5	3.9	15	62.5	30
Ciprofloxacin	5.0	20	5.0	30	5.0	20	5.0	30	5.0	14	----	-----
Imidazole											20	14

Note: Values are average of three observations.
D is diameter of zone of extract of oil and antibiotics
Z is zone of inhibition of test organisms
k.pneu is Klebsiella pneumonia,
S.aureus is Staphylococcus aureus,
P.aerugi is Pseudomonas aeruginosa,
E.coli is Escherichia coli,
S.typhi is Salmonella typhi,
C.albicans is Candida albicans

Table 3: Minimum Inhibitory Concentration (Mic) Of Cocos Nucifera Oil Extraction against Different Micro-Organisms.

MICRO-ORGANISMS	MIC DILUTION	MIC VALUE (µg/ml)
Klebsiella pneumonia	1:128	7.8
Staphylococcus aureus	1:128	7.8
Pseudomonas aeruginosa	1:16	6.3
Escherichia coli	1:32	31.3
Salmonella typhi	1:64	15.6
Candida albicans	1:64	15.6

Note: Values are average of three observations

Table 4: Diameter of Zone Of Inhibition of Cocos Nucifera Oil against Test Organisms

Extract/antimicrobial disc	k.pneu		s.aureus		p.aerugi		e.coli		s.typhi		c.albicans	
	D	Z	D	Z	D	Z	D	Z	D	Z	D	Z
Cocos nucifera oil	7.8	23	7.8	30	6.3	32	31	23	15.6	33	15.6	20
Ciprofloxacin	5.0	30	5.0	30	5.0	30	5.0	30	5.0	30	---	---
Imidazole									20	14		

Note: Values are average of three observations
D is diameter of zones of extract of oil and antibiotics
Z is zone of inhibition of test organisms
K.pneu is Klebsiella pneumonia,
S.aureus is Staphylococcus aureus,
P.aerugi is Pseudomonas aeruginosa,
E.coli is Escherichia coli,
S.typhi is Salmonella typhi,
C.albicans is Candida albicans

Table 5: Minimal Bactericidal and Minimal Fungicidal Values of Cympobogon citratus and Cocos nucifera OIL.

Micro-organisms	MBC(Cymp)	MBC(cocos)	MFC(Cymp)	MFC(cocos)
Klebsiella pneumonia	15.6	31.3		
Staphylococcus aureus	15.6	15.6		
Pseudomonas aeruginosa	31.3	12.5		
Eshericha coli	31.3	31.3		
Salmonella typhi	3.9	31.3		
Candida albicans	----		62.5	15.6

Note: Values are averages of three observations
MBC is Minimal bactericidal concentration,
MFC is Minimal fungicidal concentration
Cymp is cympobogon citratus oil,
Cocos is cocos nucifera oil

DISCUSSION

The findings from this study has distinctively shown the notable antibacterial and antifungal activity of *Cocos nucifera* oil and *Cympobogon citratus* oil when compared to ciprofloxacin and imidazole.The extract of the *Cympobogon citratus* and *Cocos nucifera* showed a varying degree of antimicrobial activity .However, *Salmonella typhi* showed the highest susceptibility to the extract of *Cympobogon citratus*.It should be noted that herbalists in south-western Nigeria has used the extract of cympobogon citratus to treat typhoid and malaria fever.(Sofowora,1993).Activities of *Cympobogon citratus* oil was found to be pronounced for *Klebsiella pneumonia* which are achieving more prominence as aetiological agents of urinary tract infection with reported isolated cases of only slightly lower than those of *Escherichia coli*(Ebie et al,2001).The extract of *Cympobogon citratus* was very effective against *Pseudomonas aeruginosa* whose resistance to most antimicrobial agent is well known(Irvin et al,1981).*Candida albicans* which has been known to be a problem for many women has been found to respond effectively to the extract of *Cympobogon citratus*.

Cocos nucifera oil has been found to be very effective against *Salmonella typhi Pseudomonas aeruginosa* and *Staphylococcus aureus* .This was in accordance with Monica, (2000) who suggested that coconut oil contains an antimicrobial agent which could make it suitable for medicinal purposes like the treatment of wound infection and urinary tract infection.

Cocos nucifera oil has also been found to be very effective against the fungi, *Candida albicans*, hence its use in the treatment of some skin diseases (Sofowora, 1996).*Cocos nucifera* oil has also been found in many body creams probably to prevent the growth of fungal skin diseases since most skin diseases are caused by fungi.

The fact that the oil extracts of *Cympobogon citratus* and *Cocos nucifera* inhibit the growth of pathogenic organisms mentioned above indicated the active components of this medicinal plants have bactericidal, bacteriostatic, and fungicidal effects supporting some of the uses in folk medicine (Sofowora, 1993).The oils of *Cympobogon citratus* and *Cocos nucifera* possess significant ($P<0.001$) antibacterial activity at very low concentration. Some antibiotics are obsolete because of the problems of drug résistance (Ekpendu et al,

1994).though there has been little attention to the antimicrobial effects of *Cympobogon citratus* and *Cocos nucifera* oils on pathogens, the consequence of drug resistance implies that new drugs both synthetic and natural must be sought to treat disease for which known drugs are no longer useful.

Further research is recommended to get the active ingredients for pharmaceutical incorporation into synthetic drugs.

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