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THE CHEMICAL AND MEDICINAL POTENTIALS OF THE FRUIT ESSENTIAL OIL OF *Chrysophyllum cainito* (INDIA STAR APPLE)

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ABSTRACT

Chrysophyllum cainito is a lesser known fruit with several medicinal applications. The essential oil was obtained by hydrodistillation and was analysed by gas chromatographys/mass spectrometry, GC/GC-MS. Twenty components were identified in the essential oil, the oil was characterized by a high proportion of fatty acids (69.37%) represented by pyruvic acid, 10-hendecenoic acid, E-9-tetradecenoic acid, pentadecanoic acid, hexadecacanoic acid and cis-9-octadecenoic acid. The oil yield was 0.19 v/w of the wet sample and its compositional profile showed markedly qualitative and quantitative variation with essential oil from Cuba. This essential oil was seen to be active against gram-positive bacteria, Escherichia coli, Klebsiella pneumoniae, Samonella typhimurium and Proteus mirabilis and also gram negative bacteria, Staphylococcus aureus and Streptococuss agalactiae.

KEYWORDS: ***Chrysophyllum cainito, essential oil, hydrodistillation, fatty acid***

RESUMO

O óleo essencial do fruto de Chrysophyllum cainito foi obtido por hidrodestilação e analisado com cromatografia gasosa e espectrometria de massa (GC-MS). Um total de vinte constituintes foram identificados no óleo essencial.que foi caracterizado pela presença alta de ácidos graxos (69.37%). Os componentes principais foram os ácidos piruvico, 10-hexadecenoico, E-9-tetradecenóico, pentadecenóico, hexadecanóico e cis-9-octadecenóico. O rendimento foi 0.19 volume/peso da amostra molhada e o perfil composicional foi muito diferente do óleo essencial de Cuba tanto qualitativamente quanto quantitativamente. O óleo essencial mostrou atividade antimicróbica tanto contra bactéria gram- positiva com Escherichia coli, Klebsiella pneumoniae, Salmonella typhimurium e Proteus mirabilis qauanto gram- negativa como Staphylococcus aureus e Streptococcus agalactiae.

PALAVRAS CHAVE:Chrysophyllum cainito, Óleo essencial, Hidrodestilação, Ácidos graxos

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INTRODUCTION

Traditional and folklore medicine plays an important role in health care services around the globe. Plants are known to synthesize a wide range of chemical substances, many of which have been and can be of tremendous value in treatment and prevention of diseases. Man has depended on the plants and plants extract as a source of medicine, food, shelter, clothing etc. since creation [15].

Herbal drugs have been one of the oldest practiced by mankind because of their cheap availability and wide application, scientist in various parts of the country concentrate on studies of these remedies in order to use these as an alternative to expensive imported drugs [12] several pharmaceutical companies are engaged in the development of natural product drugs through the isolation of the so-called active molecule from plant extracts. It is estimated that today, plant material are present in, or have provided the models for 50% western drugs [14].

Chrysophyllum cainito commonly called star apple belongs to the family sapotaceae, it usually comes in two forms, either the dark purple skinned variety with red-purple pulp or the green skinned variety with clear white pulp. In Nigeria, this fruit is not commonly eaten like the African star apple fruit, *chrysophyllum albidum.* This plant is a medicinal plant which contain substances that could be used for therapeutic purposes of which are precursors for the synthesis of useful drugs [1].

In Ivory Coast, decoction of the leaves is used for the treatment of hypertension, while in Cuba, a decoction of the leaves is used as a cancer remedy, while that of the bark is used as an antitussive (cough suppressant) [9]. In Eastern part of Africa, infusion of the leaves have been used against diabetes and articular rheumatism, while the bark is considered a tonic and stimulant. In Venezuela, the fruit and the seed are used as a diurectic and the ripe fruits because of their mucilaginous character are eaten to sooth inflammation, laryngitis and pneumonia. The latex of the tree in Brazil is applied on abscesses [7]. In Nigeria, few people eat the fruit because it is not common but those that eat it do so because of the sweetness and its mucilaginous character.

Chrysophyllum cainito belongs to the family *sopotaceae* and their chemical and biological studies are scarce [2] and there is lack of chemical information about the genus *chrysophyllum,* but a number of reports concerning the isolation and characterization of bioactive compounds from various parts of the plant have appeared in the literature.

The volatile constituents of star apple were extracted in Cuba [10], the essential oil were analyzed by GC-MS and one hundred and four compounds were identified in the aroma concentrate of which *(E) -2-hexanal, limonene, linalool, copaene and hexadecanoic acid* were found to be the major constituents.

In another study from Florida, fresh fruit of chrysophyllum cainito were extracted with methanol and partitioned with hexane and ethyl acetate sequentially. Nine known polyphenolic antioxidants were identified and epicatechin is present in highest concentration [6].

The aqueous extract of the leaves was found at a dose of 20g/l to reduce the hyperglycemia of diabetic rat from 5g/l to 1.4g/l [5].

The extract of *chrysophyllum cainito* was used in goats against *Haemonchos contostus*[4] it was reported that the extract has nematocidal activity and this confirmed its use in ethnoveterinary practices and animal health management [11].

The purpose of the present investigation therefore was to extract, analyse and determine the antimicrobial activity of the essential oil of this lesser known fruit of *chrysophyllum cainito*.

MATERIALS AND METHODS

Plant material and essential oil isolation

Fresh fruits of *chrysophyllum cainito* (dark purple skinned variety) were collected from the campus of Ajayi Crowther University, Oyo in South West Nigeria in March, 2013. The sample was authenticated at the Forestry Research Institute of Nigeria (FRIN), Ibadan, where voucher specimens were deposited and was given voucher number FHI 109475. A sample of 700g was pulverized using mortar and pestle. The fresh fruit was subjected to hydro-distillation method using an all glass Clevenger apparatus according to procedure described by European Pharmacopoeia [18]. The oil was collected and kept in refrigerator without further treatment until GC/GC-MS analysis.

Gas Chromatography/GC mass spectrometric analysis

The chemical composition of the essential oil was analysed using GC-MS technique. The mass spectrometer was SHIMADZU GCMS-QP2010 plus (Shimadzu Corporation, Japan) in the electron impact (EI) ionization mode (70ev) and HP 5MS (bonded 0.25μm capillary column (restek, Bellefonte, PA) Injector and detector temperature was held at 60°C for 30 minutes then programmed to 240°C at rate of 5°C/min. Helium (99.99%) was the carrier gas at a flow rate of 1ml/min. Diluted samples (1/100 in hexane v/v) of 1.0ml were injected automatically, the linear velocity of the column was 36.8cm/sec, each peak was then analysed and assigned a number in the order that it was detected. The identification of the components was based on comparison of their mass spectra with those of NIST library, mass spectra database and literature.

Antimicrobial Activity Tests

The antibacterial activities of the essential oil was measured against gram-negative (*Escherichia coli, Klebsiella pneumoniae, Salmonella typhimurium and Proteus mirabelis* and gram-positive *Staphylococcus aureus and Streptococcus agalactiae* using a well diffusion method according to the National Committee for Clinical Laboratory Standard [8]. Briefly, Petri plates containing approximately 25-30ml of nutrient agar medium were swabbed using cotton applicator with a 24 hours sub-cultured bacterial strains which were prepared in dilution to match the turbidity intensity of the MacFarland Standard. Wells (6mm diameter) were punched in the agar and filled with 10μl of the extracts of different concentrations (1000, 100 and 10$\mu g ml^{-1}$). The plates were incubated at 37°C for 24 hours. The antibacterial activities were assessed by measuring the inhibition zone diameter (mm) around the well.

RESULTS AND DISCUSSION

Hydro distillation of the fresh fruit of *chrysophyllum cainito* produced a clear light essential oil. The oil yield is 0.19% v/w of the wet sample. The chemical components identified by GC/GC-MS are listed in Table 1. It was characterized by a high proportion of fatty acids (69.37%) represented by (IIz) –II hexadecenoic acid (24.42%), cis-9-octadecenoic acid (oleic acid) (13.04%), E-9- Tetradecenoic (12.21%), E-9-Hexadecanoic acid (9.54%), Pentadecanoic acid (3.18%), Nonanoic acid (pelargic acid) (3.18%), Pyruvic acid (2.97%) and 10-Hendecenoic acid (0.83%).

The compositional profile of the fruit oil showed markedly qualitative and quantitative variation with that from Cuba. *(E)-2-hexanal, limonene, linalool and copaene* which are part of the major constituents of the oil from Cuba were not identified from the Nigerian oil sample, but hexadecanoic acid which was reported as one of the major constituents [10] is present in the Nigerian sample (9.54%).

The oil is seen at different concentration to be active against gram-positive bacteria (K*lebsiella pneumoniae, Salmonella typhimurium and Proteus mirabilis* and also gram-negative bacteria *Staphylococcus aureus and Streptococcus agalactiae* as listed in Tables 2 and 3 and this can be compared with standard antibiotics as listed in Tables 4 and 5.

Table 1. Chemical Constituents of the fruit essential oil of *chrysophyllum cainito*

S/N	COMPONENT	RI	% COMPOSITION
1	3-ethoxy-1-butane	620	0.80
2	Divinyl sulfide	650	2.97
3	2,2-Dimethylpentanal	821	0.80
4	2-pentanethiol	837	2.97
5	Heptan-3-one (ethylbutylketone)	853	0.80
6	Tert-Butylcarbinol	876	0.80
7	Pyruvic acid	919	2.97
8	Monochloromethylisopentanoate	946	0.80
9	Allyl pentanoate	974	2.97
10	Methylcyclooctane	1020	0.83
11	2(3H)-Furanone	1061	2.97
12	(4Z)-4-Methyl-4-undecene	1199	0.83
13	Nonanoic acid (pelargic acid)	1272	3.18
14	10-Hendecenoic acid (Sevinon)	1461	0.0
15	E-9-Tetradecenoic acid	1777	12.21
16	Pentadecanoic acid (pentadecyclic acid)	1869	3.18
17	Hexadecanoic acid (palmitic acid)	1968	9.54
18	(11z)-11-Hexadecenoic acid	1976	24.42
19	E-2-Octadecadecen-1-0l	2061	12.21
20	Cis-9-octadecenoic acid (oleic acid)	2175	13.04
21	Methyl(13E)-13-docosenoate	2483	0.83
	Total		99.95%

Table 2.

Zones of inhibition (mm) showing the antimicrobial activities of the essential oil at different concentrations (Gram-negative bacteria)

Organism	*Escherichia coli*	*Klebsiella pneumoniae*	*Salmonella typhimorium*	*Proteus mirabilis*
Conc (μgml^{-1})	1000 100 10	1000 100 10	1000 100 10	1000 100 10
Zone of inhibition	15 08 09	13 13 13	12 12 12	13 13 13

Keynote –= no inhibition, 6-9mm = low inhibition 10-15mm = moderate inhibition≥15mm = high inhibition.

Table 3.

Zones of inhibition (mm) showing the antimicrobial activities of the essential oil at different concentrations(Gram-positive bacteria).

Organism	Staphyococcus aureus	Streptococcus agalatiae
Conc (μgml^{-1})	1000 100 10	1000 100 10
Zone of inhibition	17 13 13	15 15 15

Keynote –= no inhibition, 6-9mm = low inhibition 10-15mm = moderate inhibition≥15mm = high inhibition.

Table 4.

Zones of inhibition (mm) showing the antimicrobial activity of antibiotics (Gram –negative bacteria)

Organism / Antibiotics/ concentrations	*Escherichia coli*	*Klebsiella pneumoniae*	*Salmonella typhimorium*	*Proteus mirabilis*
Nitrofurantoin (200μg)	25	20	15	15
Ofloxacin (5μg)	20	20	22	33
Gentamicin (10μg)	24	15	18	16

Keynote –= no inhibition, 6-9mm = low inhibition 10-15mm = moderate inhibition≥15mm = high inhibition.

Table 5.

Zones of inhibition (mm) showing the antimicrobial activities of antibiotics (Gram –positive bacteria)

Organism / Antibiotics/ concentrations	*Staphylococcus Aureus*	*Streptococous agalactiae*
Nitrofurantoin (200μg)	28	11
Ofloxacin (5μg)	20	-
Gentamicin (10μg)	-	-

Keynote –= no inhibition, 6-9mm = low inhibition 10-15mm = moderate inhibition≥15mm = high inhibition.

CONCLUSION

The results obtained in this study showed that the fruit essential oil of *chrysophyllum cainito* from South West Nigeria has a qualitative and quantitative variation from the one obtained in Cuba and this can be traced to the tropical nature of Nigeria. This study is the first in Nigeria, to the best of my knowledge.

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