Microbiology Research International
Vol. 3(3), pp. 41-50, August 2015
ISSN: 2354-2128
Full Length Research Paper

Antimicrobial activities and chemical compositions of *Chrysophyllum cainito* (star apple) fruit

S. U. Oranusi, W. Braide* and R. U. Umeze

Department of Microbiology, Federal University of Technology Owerri, P.M.B 1526, Owerri, Imo State, Nigeria.

Accepted 10 August, 2015

ABSTRACT

The pulp and seed of *Chrysophyllum cainito* was analyzed for its antimicrobial potentials, microbial profile, anti-nutrients and nutrients using standard methods. The pulp and seed showed varying levels of antimicrobial activities against some clinical isolates such as *Escherichia coli* and species of *Salmonella*, *Staphylococcus*, *Pseudomonas*, *Aspergillus*, *Candida* and *Penicillium*. The microbial count of the fruits ranged from 1.0×10^9 to 2.4×10^{10} for total aerobic plate count, 1.0×10^7 to 2.0×10^7 for fungal count and 1.0×10^8 to 1.2×10^9 for coliform count. The identities of the normal flora on the surfaces and pulp of healthy fruits and of spoilage organisms were confirmed to include species of *Bacillus*, *Corynebacterium*, *Staphylococcus*, *Micrococcus*, *Acinetobacter*, *Enterococcus*, and *Pseudomonas.* The fungal isolates include species of *Rhizopus*, *Aspergillus*, *Penicillum* and *Saccharomyces*. The high microbial counts and their presence, portends serious health implication. *Aspergillus* and *Penicillium* species produces mycotoxins involved in mycotoxicosis of humans and animals. *Staphylococcus* and *Bacillus* species produce potent toxins implicated in food borne illnesses, while the presence of *Enterococcus* indicates feacal contamination. Varying concentrations of phytochemicals such as saponin, flavonoids, tannin, steroid and cardiac glycoside were detected. Vitamins such as vitamin A (0.027 to 0.089 mg) and vitamin C (10.00 to 43.54 mg) were also present. Minerals such as calcium, magnesium, phosphorus, potassium and sodium were present at concentrations of 37.0, 5.0, 8.0, 38.0 and 21.0 mg respectively, for the pulp extracts. The proximate composition of star apple consists of protein (1.96 to 4.63 g), moisture (56.04 to 75.90 g), fat (0.88 to 15.81 g), fibre (2.31 to 4.19 g), ash (0.56 to 0.84 g) and carbohydrate (18.39 to 78.49 g). *Chrysophyllum cainito* holds great potentials as an antimicrobial agent for chemotherapeutic medicine and it is a rich source of nutrient and phytochemicals.

Keywords: *Chrysophyllum cainito,* anti-nutritive composition, antimicrobial activity.

*Corresponding author. E-mail: amakam4xrist@yahoo.co.uk, wesleybraide2005@yahoo.com. Tel: +234-0803-7100-964.

INTRODUCTION

Fruits have been traditionally and nutritionally important food to man for many years (Adepoju and Adeniji, 2012). Fruits are part of plants that produces the seed that are edible and also provide nutrients to man (Adepoju and Adeniji, 2012). The chemical compositions and the antimicrobial sensitivity of some fruits such as *Chrysophyllum albidum* (udara) and *Persea americana* (Avocado pear) have been determined (Arukwe et al., 2012). Fruits have been known to be medicinal and have been applied in curative medicine.

Chrysophyllum cainito also known as star apple is a fleshy fruit with soft endocarp. It is a tropical tree of the family Sapotaceae, and native to the Greater Antilles and the West Indies (Luo et al., 2002). It is also sparsely distributed in Nigeria; grows rapidly and reaches 20 m in height (Luo et al., 2002). It has numerous common names including cainito, caimito, star apple, golden leaf tree and also milk fruit (National Research Council, 2008). The tree is hermaphrotic in nature with round; purple-skinned fruit that is often green around the calyx with a star pattern in the pulp. Sometimes there is a greenish – white or yellow variety of the fruit (National Research Council, 2008). The skin is rich in latex; though the skin and the rind are not edible. The seed is flat, hard

and light brown in color (Einbond et al., 2004). The fruits are delicious as a fresh dessert fruit; it is sweet and best served chilled. The fruit has antioxidant properties (Luo et al., 2002; Einbond et al., 2004). Infusions of the leaves have been used against diabetes and articular rheumatism (Luo et al., 2002). The bark is considered a tonic and stimulant, and a bark decoction is used as an antitussive (Luo et al., 2002).

The Ayurvedic system of medicine has described various fruits in the treatment of diseases, which play an important role in modern health care and curing various ailments and diseases (Luo et al., 2002). There are several reports on the chemical composition and antimicrobial activities of some fruits and their extracts that inhibits various bacteria (Luo et al., 2002). However, studies on the chemical compositions and antimicrobial activities of *Chrysophyllum cainito* are limited. Therefore, scientific evaluation of this fruit is important to elucidate its chemical composition as well as its antimicrobial activity in order to support its use as food and as alternative medicine in the treatment of some infections, especially enteric diseases.

This study reports on the antimicrobial potential of the fruit and its nutritional and phytochemical properties.

MATERIALS AND METHODS

Sample collection

One hundred fresh and well ripened star apples were harvested from the tree located within the premises of Federal University of Technology, Owerri (F.U.T.O), Imo state, Nigeria between the months of February and March, 2013.

Antimicrobial assay of the pulp and seed of *Chrysophyllum cainito*

Pure cultures of multiple drug resistance *Staphylococcus aureus*, *Escherichia coli*, *Salmonella typhi*, *Pseudomonas aeruginosa*, *Candida albicans, Aspergillus* and *Penicillium* were obtained from the microbiology unit in the Federal Medical Center, Owerri, Imo State, Nigeria. The identities of the cultures were confirmed using routine laboratory methods (Sharma, 2007), maintained on agar slant at 4°C prior to use.

The antimicrobial activity was performed by agar well diffusion methods (Dalitha, 2008). The antimicrobial diffusion test was carried out using a cell suspension obtained from McFarland turbidity standard number 0.5. Antimicrobial activity was evaluated by measuring the diameter of inhibition zone around the extract.

Minimum inhibitory concentration (MIC) test

Minimum inhibitory concentrations of the extracts (seed and pulp) were determined by two-folds serial dilution method (Chandrasekaran and Venkatesalu, 2004). The dose levels of 250.00, 125.00, 62.50, 31.25 mg/L and 15.62 mg/ml concentrations each of the extracts were used for MIC determination. After suitable incubation (at 37°C for 24 h), the least concentration of the sample extracts with no visible growth was taken as the MIC. This was further confirmed spectrophotometrically at an optical density (OD) of 520 nm.

Minimum bacteriocidal concentration (MBC)

A sample was taken from the tubes with no visible growth in the MIC test and plated out according to the method of Banso and Adeyemo (2007) and incubated at 37°C for 24 h. After the incubation, the plates were observed for growth. The lowest concentration of the extracts without any growth was noted as the minimum bactericidal concentration (MBC) in mg/ml.

Microbiological analysis of the external surface of the healthy fruit and pulp

The fruit was aseptically washed with 100 ml normal saline into a sterile beaker and serial dilution was made by transferring 1 ml portion until 10 fold serial dilution was obtained. Aliquot portion (0.1 ml) was inoculated onto bacteriological and mycological media. The inocula were spread evenly to ensure countable colonies, and incubated at 37°C for 24 h (Cheesbrough, 2000).

Twenty grams of the pulp was weighed and aseptically introduced into a sterile stomacher blender containing 180 ml of normal saline and agitated to mix thoroughly. A portion (1 ml) was aseptically transferred into 9 ml normal saline and further diluted decimally. An aliquot portion (0.1 ml) was inoculated into the media, spread evenly and incubated at 37°C for 24 h (Cheesbrough, 2000; Sharma, 2007).

Microbiological analysis of deteriorated pulp and outer skin surface of the fruit

This was done using the method applied for healthy fruit, according to Cheesbrough (2000) and Sharma (2007).

Enumeration and identification of microorganisms

Total count of bacterial and fungal isolates were done using Gallemkamp digital colony counter and hand lens respectively (Harrigan and McCance, 1990; Sharma, 2007), and expressed as colony forming units per grams/milliliters (cfu/g/ml). The identity of the bacterial isolates was done on the basis of their colonial, microscopic and biochemical characteristics (Beishir, 1987; Cheesbrough, 2000; Pelczar et al., 1993; Sharma, 2007). The identity of the fungal isolates was done based on their mycelial arrangement, sporulation, pigmentation on reverse and surface of media (Domsch et al., 1993). The identities of bacteria and fungi isolated were confirmed with reference to standard identification atlas and keys (Buchannan and Gibbon, 2000; Barnett and Hunters, 1987).

Determination of anti-nutritional components

Chemical tests were carried out on the pulp and seed extracts using standard procedures to identify the constituents as described by Denwick (2002), Buhler (2000), George et al. (2002) and Heslem (1989). Active constituents analyzed include alkaloid, anthraquinone, steroid, flavonoid, cardiac glycoside, tannin and saponin.

Determination of mineral contents of the pulp

Potassium and sodium were determined using Flame Photometer

Table 1. Antimicrobial activity of pulp extracts showing the mean total of 50 isolates with their zones of inhibition.

Pathogens	Pulp extract [Zone of inhibition (mm)]	Antibiotic [Zone of inhibition (mm)]	Standard for zone of inhibition for commercial antibiotics (mm)
		Gentamicin	Gentamicin
Salmonella spp.	1	19	19-25
Staphylococcus spp.	6	18	17-28
Escherichia coli	5	19	18-26
Pseudomonas spp.	10	17	16-21
		Ketoconazole	Ketoconazole
Aspergillus spp.	2	16	14-16
Penicillium spp.	1	19	19-21
Candida spp.	3	20	20-22

Source for the Standards: Erhabor et al. (2013).

method (Bonire et al., 1990). Phosphorus was determined by Vanado-molybdate colorimetric method (Ologhobo and Fetuga, 1983). Calcium and magnesium was determined by complexiometric method using EDTA (Bonire et al., 1990).

Analysis of vitamins

Vitamin C was determined by visual titration method (Ness et al., 1996). Vitamin A was determined using B-carotene estimation method (Kirk and Sawyer, 1998).

Nutritional evaluation of the pulp

Moisture analysis was determined by gravimetric method (AOAC, 2000). Crude protein was determined by Kjedahl method (Chang, 2003) in which the nitrogen content was determined and multiplied by 6.25 to obtain the protein content. Ash content was determined by furnace incineration gravimetric method (James, 1995). Soxhlet solvent extraction method by James (1995) was employed in the determination of fat content. Crude fibre was determined by the Weende gravimetric (AOAC, 2000).

RESULTS

Tables 1 and 2 show the antimicrobial activity results of the pulp and seed extracts, respectively tested against fifty isolates each. Gentamicin and Ketoconazole were used as control drugs.

The zones of inhibitions (ZOI) of the different concentrations of the pulp extract of *Chrysophyllum cainito* measured in millimeter (mm) are represented in Table 3. Antimicrobial activity is high against *Staphylococcus*, *Pseudomonas* and *Salmonella* at concentrations 250.00, 250.00 and 31.25 mg/ml respectively.

The zone of inhibition measured in millimeter (mm), of the seed extract of *C. cainito* is shown in Table 4. Antimicrobial activity is prominent at the higher concentrations (250 and 125 mg/ml). The seed and pulp extracts exhibited higher antimicrobial activity against *E. coli* and *Staphylococcus aureus* than other test organisms including the fungi.

The minimum inhibitory concentration of various concentrations of pulp and seed extract taken at optical density (OD) at 520 nm is shown in Tables 5 and 6 respectively. Optical density (OD) was measured with an ultra violet spectrophotometer.

The Minimum Bactericidal Concentration (MBC) of the various MIC concentrations of both the pulp and seed extract showed scanty growths and in some cases, no growth at all. These are shown in Table 7.

The mean total microbial count is shown in Table 8. The total aerobic plate count ranges from 1.0×10^9 to 2.4×10^{10}, the coliform count ranged from 1.0×10^8 to 1.2×10^9 with the fungal count ranging from 1.0×10^7 to $2.0 \times 10^{7.}$

The microorganisms (fungi and bacteria) isolated from both healthy and the deteriorated fruits are shown in Table 9. Species of *Bacillus*, *Staphylococcus*, *Pseudomonas, Enterococcus*, *Micrococcus*, *Corynebacterium*, *Acinetobacter*, *Penicillium*, *Fusarium*, *Saccharomyces* and *Rhizopus* were isolated.

Qualitative phytochemical analysis shows the presence of saponins, tannins, flavonoids, steroids, cardiac glycosides and anthraquinones (Table 10).

Table 11 showed the vitamin and mineral compositions of the fruit extract. Vitamins analyzed include; vitamin A and C with the pulp extract containing a high amount of vitamin C than the seed extract. The minerals analyzed included calcium, magnesium, phosphorous, potassium and sodium.

Table 12 shows the result of the quantitative values of moisture, carbohydrate, fat, protein, fibre and ash, as well as the total energy.

Table 13 shows the nutritional content and mineral compositions of the pulp and seed extracts of the fruit (*C. cainito*) as compared with standard values of the Reference Nutrient Intake (RNI) from international agency. The gross energy of the seed is higher than that

Table 2. Antimicrobial activity of seed extracts showing the mean total of 50 isolates with their zones of inhibition.

Pathogens	Seed extract [Zone of inhibition (mm)]	Antibiotic [Zone of inhibition (mm)]	Standard for zone of inhibition for commercial antibiotics (mm)
		Gentamicin	Gentamicin
Salmonella spp.	1	19	19-25
Staphylococcus spp.	8	18	17-28
Escherichia coli	10	19	18-26
Pseudomonas spp.	2	17	16-21
		Ketoconazole	Ketoconazole
Candida spp.	1	16	14-16
Penicillium spp.	2	19	19-21
Aspergillus spp.	0	20	20-22

Source for the Standards: Erhabor et al. (2013).

Table 3. Zones of inhibition of various concentrations of the pulp extract of *Chrysophyllum cainito.*

Pathogens	Conc. of pulp extract (mg/ml)	Z0I (mm)
Staphylococcus spp.	250	6
Staphylococcus spp.	125	4
Staphylococcus spp.	62.5	1
Staphylococcus spp.	31.25	1
Staphylococcus spp.	15.62	1
Salmonella spp.	250	1
Salmonella spp.	125	1
Salmonella spp.	62.5	3
Salmonella spp.	31.25	9
Salmonella spp.	15.62	1
E.coli	250	5
E.coli	125	1
E.coli	62.5	1
E.coli	31.25	1
E.coli	15.62	5
Pseudomonas spp.	250	10
Pseudomonas spp.	125	0
Pseudomonas spp.	62.5	1
Pseudomonas spp.	31.25	1
Pseudomonas spp.	15.62	1
Candida spp.	250	3
Candida spp.	125	1
Candida spp.	62.5	1
Candida spp.	31.25	1
Candida spp.	15.62	1
Aspergillus spp.	250	2
Aspergillus spp.	125	1
Aspergillus spp.	62.5	1
Aspergillus spp.	31.25	1
Aspergillus spp.	15.62	4

Table 3. Continues.

Penicillium spp.	250	1
Penicillium spp.	125	1
Penicillium spp.	62.5	1
Penicillium spp.	31.25	1
Penicillium spp.	15.62	2

Key: Z0I = Zone of Inhibition. mm = millimeter.

Table 4. Zones of inhibition of various concentrations of seed extract of *Chrysophyllum cainito.*

Pathogens	Conc. of pulp extract (mg/ml)	Z0I (mm)
Staphylococcus spp.	250	8
Staphylococcus spp.	125	6
Staphylococcus spp.	62.5	4
Staphylococcus spp.	31.25	2
Staphylococcus spp.	15.62	0
Salmonella spp.	250	1
Salmonella spp.	125	1
Salmonella spp.	62.5	1
Salmonella spp.	31.25	1
Salmonella spp.	15.62	0
E.coli	250	10
E.coli	125	6
E.coli	62.5	3
E.coli	31.25	2
E.coli	15.62	2
Pseudomonas spp.	250	2
Pseudomonas spp.	125	2
Pseudomonas spp.	62.5	1
Pseudomonas spp.	31.25	1
Pseudomonas spp.	15.62	1
Candida spp.	250	1
Candida spp.	125	1
Candida spp.	62.5	0
Candida spp.	31.25	2
Candida spp.	15.62	1
Aspergillus spp.	250	2
Aspergillus spp.	125	1
Aspergillus spp.	62.5	1
Aspergillus spp.	31.25	1
Aspergillus spp.	15.62	1
Penicillium spp.	250	0
Penicillium spp.	125	1
Penicillium spp.	62.5	3
Penicillium spp.	31.25	1
Penicillium spp.	15.62	1

Key: Z0I = Zone of Inhibition; mm = millimeter.

Table 5. Minimum inhibitory concentration (MIC) of various concentrations of pulp extract, OD. measured by U.V. spectrophotometer (OD: 520 nm).

Conc. of seed extract (mg/L)	Pulp extract against *E.coli*	Pulp extract against *Staph. spp*	Pulp extract against *Salm. spp*	Pulp extract against *Aspergillus spp*	Pulp extract against *Ps. spp*	Pulp extract against *Candida spp*	Pulp extract against *Pencillium spp*
250	**0.359±0.03**	1.890±0.07	1.821±0.05	1.568±0.01	1.839±0.003	1.575±0.02	1.911±0.01
125	0.511±0.08	1.181±0.02	0.766±0.13	0.818±0.01	0.946±0.003	0.621±0.04	1.005±0.01
62.5	0.416±0.05	0.537±0.06	0.780±0.09	0.336±0.004	0.611±0.005	0.351±0.02	**0.652±0.01**
31.25	1.652±0.02	**0.356±0.03**	0.665±0.04	**0.092±0.01**	0.514±0.003	0.096±0.02	0.881±0.02
15.62	0.648±0.02	0.517±0.06	**0.627±0.01**	0.142±0.004	**0.449±0.004**	**0.090±0.02**	0.669±0.01

Table 6. Minimum inhibitory concentration (MIC) of various concentrations of seed extract, OD measured by U.V. spectrophotometer (OD: 520 nm).

Conc. of seed extract (mg/L)	Seed extract against *E. coli*	Seed extract against *Salmonella* spp	Seed extract against *Staph*.spp	Seed extract against *Ps*. spp	Seed extract against *Candida* spp	Seed extract against *Aspergillus* spp	Seed extract against *Penicillium* spp
250	3.000±0.36	3.000±0.36	3.000±0.36	3.000±0.36	3.000±0.39	2.052±0.01	2.210±0.02
125	2.305 ±0.13	2.523±0.18	2.561±0.19	2.612±0.002	2.646±0.03	1.374±0.01	1.480±0.02
62.5	1.863±0.12	2.015±0.14	1.848±0.07	1.778±0.004	1.444±0.03	0.739±0.01	0.845±0.02
31.25	**1.509±0.07**	**1.576±0.03**	1.680±0.06	**1.602±0.003**	1.618±0.03	**0.144±0.004**	0.657±0.03
15.62	1.584±0.03	1.658±0.02	**1.664±0.16**	1.611±0.002	**1.364±0.01**	0.150±0.004	**0.614±0.01**

Table 7. Minimum bactericidal concentration (MBC) of seed and pulp extracts of *Chrysophyllum cainito.*

Organisms	At all concentrations (mg/ml)	
	Pulp	Seed
Escherichia coli	Very scanty growth	Scanty growth
Staphylococcus spp.	No growth	Scanty growth
Salmonella spp.	No growth	Scanty growth
Candida spp.	No growth	No growth
Pseudomonas spp.	Very scanty growth	Scanty growth
Aspergillus spp.	Scanty growth	No growth
Penicillium spp.	No growth	Very scanty growth

Table 8: Mean total microbial count (cfu/g).

Sample	Total aerobic plate count	Coliform count	Fungal count
A1	1.1×10^{10}	1.0×10^{8}	1.1×10^{8}
B1	1.0×10^{9}	1.0×10^{8}	1.0×10^{8}
C1	1.2×10^{9}	1.0×10^{8}	1.0×10^{7}
D1	1.2×10^{10}	1.0×10^{8}	1.0×10^{7}
E1	1.3×10^{10}	1.1×10^{10}	1.2×10^{8}
A2	1.0×10^{9}	1.0×10^{8}	1.3×10^{8}
B2	1.1×10^{9}	1.0×10^{8}	1.3×10^{8}
C2	2.0×10^{9}	1.0×10^{8}	1.0×10^{7}
D2	1.0×10^{9}	1.0×10^{8}	1.0×10^{7}
E2	2.4×10^{10}	1.2×10^{9}	2.0×10^{7}

Key: A1-D1= Healthy pulp; E1 = Deteriorated pulp; A2-D2= Healthy fruit surface; E2 = Deteriorated fruit surface.

Table 9. Microbial isolates from samples.

Sample	Microbial isolates
Healthy fruit	*Bacillus* spp., *Staphylococcus aureus*, *Micrococcus* spp., *Corynebacterium* spp., *Acinetobacter* spp., *Rhizopus* spp., *Aspergillus* spp., *Penicillum* spp. and *Saccharomyces* spp. and *Enterococcus* spp
Deteriorated fruit	*Bacillus* spp., *Staphylococcus aureus*, *Micrococcus* spp., *Corynebacterium* spp., *Acinetobacter* spp., *Pseudomonas* spp., *Enterococcus* spp., *Rhizopus* spp., *Fusarium* spp., *Penicillum* spp. and *Saccharomyces* spp.

Table 10. Phytochemical composition of seed and pulp extracts.

Sample	Saponin	Tannin	Alkaloid	Flavonoid	Steroid	Cardiac glycoside	Anthraquinone
Pulp	+++	+++	+++	-	+++	+++	-
Seed	-	+	-	+++	+	-	-

Key: +++ (Highly concentrated); + (Trace); - (Absent).

Table 11. Mineral and vitamin composition of the seed and pulp of *Chrysophyllum cainito.*

Parameter	Pulp	Seed
Vit.C (mg/100 g)	43.54 ± 0.57	10.0 ± 0.22
Vit.A (mg/100 g)	0.089 ± 0.32	0.027 ± 0.44
Calcium (mg/100 g)	37.0 ± 0.25	168.0 ± 0.28
Magnesium (mg/100 g)	5.0 ± 0.24	90.0 ± 0.25
Phosphorous (mg/100 g)	8.0 ± 0.25	18.0 ± 0.31
Potassium (mg/100 g)	38.0 ± 0.29	78.0 ± 0.32
Sodium (mg/100 g)	21.0 ± 0.32	39.0 ± 0.25

Table 12. Total energy and proximate composition (%) of the pulp and seed extract.

Sample	Total energy (Kcal/100g)	Moisture	Fat	Fibre	Protein	Ash	Carbohydrate
Seed	474.77±1.09	56.04±1.64	15.81±0.53	4.19±0.46	4.63± 0.12	0.84±0.25	78.49±0.40
Pulp	89.32±0.15	75.90±4.31	0.88±0.31	2.31±0.21	1.96±0.23	0.56±0.10	18.39±0.33

Table 13. Nutrient composition of *C. cainito* (seed and pulp) compared with the reference nutrient intake (RNI).

Nutrient	Nutrient of pulp (100 g)	Nutrient content of seed extract (100 g)	RNI
Gross Energy (kcal)	89.32	474.77	2300
Protein (g)	1.96	4.63	50
Fibre (g)	2.31	4.19	20
Potassium (mg)	38.0	78.0	3500
Calcium (mg)	37.0	168.0	400
Magnesium (mg)	5.0	90.0	270
Phosphorous (mg)	8.0	18.0	550
Vitamin C (mg)	43.54	10.0	60
Vitamin A (µg)	89.0	27.0	625
Sodium (mg)	21.0	39.0	1600

RNI source: www. nap.edu.

of the pulp, whereas the pulp shows higher vitamin C (ascorbic acid) than the seed.

DISCUSSION

This study shows that *C. cainito* has great potentials as antimicrobial agents against selected pathogens and may be used as an alternative medicine in the treatment or control of enteric bacterial infections. The results also showed that the test organisms were potentially susceptible to the seed and pulp extracts based on their zones of inhibition which ranged from 1mm to 10mm (Tables 3 and 4). The results of the antimicrobial susceptibility assay, the MIC assays and the MBC assay, showed promising evidence for the antimicrobial activity of *C. cainito* pulp and seed extracts against enteric pathogens.

The mean total microbial count of the samples were analyzed and the result shown in Table 8. Most of the bacteria and fungi isolated from the sample are widely distributed in nature, particularly in the soil and may contaminate the samples during harvest and handling. The microbial isolates found on the surfaces and pulp of the healthy and deteriorated fruit is shown in Table 9. These isolates which includes species of *Bacillus*, *Staphylococcus*, *Micrococcus*, *Corynebacterium*, *Acinetobacter*, *Enterococcus*, *Pseudomonas*, *Rhizopus*, *Aspergillus*, *Penicillum*, *Fusarium* and *Saccharomyces* are pathogenic microorganisms whose natural habitats includes the human intestinal tracts, respiratory tracts and or droplets, skin, soil and water. Ingestion of these pathogens followed by their growth and tissue invasion and or release of toxins may cause food borne infection (Hoge et al., 1991). Therefore it is important that fruits be properly handled and washed before consumption.

Phytochemicals are important chemicals found virtually in plants at different concentrations (Duke, 1992). Table 10 shows the result of phytochemicals analyzed. It shows that the pulp extract contained a high concentration of saponin while it is absent in the seed extract. General characteristics of saponins includes formation of foams in aqueous solution, haemolytic activity, cholesterol - binding properties, etc (Sodipo and Akiniyi, 2000). Saponins also confer antimicrobial effects. Possible antimicrobial mechanism of saponins is as a result of reduced glucose utilization efficiency in microorganism, thus affecting their growth and proliferation; reducing the activity of key enzymes in physiological metabolism and suppressing the synthesis of relevant proteins and finally executing the antimicrobial effect (Yu et al., 2013). Tannins noted for astringency and bitter taste, hasten the healing of wounds and inflamed mucus membrane (Duke, 1992). The result on Table 10 showed that the pulp extract contains a high concentration of tannin, while the seed contains just a trace amount of tannin. Tannic acid was found to be inhibitory to the growth of intestinal bacteria such as *Bacteroides fragilis*, *Clostridium perfringens*, *Escherichia coli* and *Enterobacter cloacae* amongst others. Tannic acid may also work like a siderophore to chelate iron from the medium and make iron unavailable to microorganisms, especially those growing under aerobic conditions that need iron for variety of functions like reduction of ribonucleotide precursor of DNA, formation of haem and other essential purposes (Chung et al., 1998). The different mechanisms proposed so far to explain tannin antimicrobial activity includes inhibition of extracellular microbial enzymes, toxic action on the membrane of the microorganisms, deprivation of the substrates required for microbial growth or direct action on microbial metabolism through inhibition of oxidative phosphorylation. A further mechanism involving iron deprivation had been proposed (Chung et al., 1998). Tannin is noted to have no antifungal activity due to their (fungi) morphological structure. Fungi have thicker cell walls and contain higher percentage of chitin (Madigan and Martinko, 2006). Flavonoids are potent water-soluble super antioxidants and free radical scavengers. They prevent oxidative cell damage, have strong anti – cancer activity and protect against all stages of carcinogenesis (Salah et al., 1995). Also, flavonoids in intestinal tract lower the risk of heart disease, inflammation and represent the most common and widely distributed groups of plant phenolic compounds. Both the pulp and seed extracts contains high levels of flavonoid and this could be responsible for the anti-inflammatory, anti-cancer and anti-hypertensive properties of the plant and its parts. Catechins (flavonoids in fruits) have been shown to inactivate cholera toxin in *Vibrio cholerae* and inhibit isolated bacterial glucosyltransferases in *Streptococcus mutans,* probably due to complexing activities (Borris, 1996). Also, the correlation between antibacterial activity and membrane interference supports the theory that flavonoids may demonstrate antibacterial activity by reducing membrane fluidity of bacterial cells (Nakahara and Kawabata, 1993). Flavonoids (myricetin, datiscetin, kaempferol and quercetin) exhibit an inhibitory activity against methicillin-resistant *Staphylococcus aureus* (MRSA). Myricetin was also found to inhibit the growth of multidrug resistant *Burkholderia cepacia*, Vancomycin- resistant Enterococci (VRE) and other medically important organisms such as *Klebsiella pneumonia* and *Staphylococcus epidermidis* (Xu and Lee, 2001). Their mode of antimicrobial action may be related to their ability to inactivate microbial adhesins, enzymes, cell envelope, transport proteins, etc. (Cowan, 1999).

Steroids were found to be present in the pulp and seed extracts, but with a higher concentration in the pulp extract. Steroidal compounds are of importance and so interest as grown in pharmacy due to their association with such compounds as sex hormones (Okwu, 2001). Since the pulp of *C. cainito* contains a high concentration of steroids, its consumption should be encouraged to

serve as potent starting materials in the synthesis of sex hormones and also to boost existing sex hormones. Table 10 also shows that the seed extract contains no cardiac glycoside, while the pulp contains a high concentration of cardiac glycoside. Cardiac glycosides help the heart to beat more efficiently by making the blood get more oxygen and gives nutrients to the body cells. Cardiac glycosides also help in the treatment of congestive heart failure and arrhythmia (Sodipo and Akiniyi, 2000). Alkaloid and anthraquinone were not found in the extracts of both seed and pulp.

Its decent amount of ascorbic acid (43.54 mg) as shown in Table 11 makes it tolerable for people with peptic ulcer. *C. cainito* is also believed to be a good source of antioxidants (β-carotene and ascorbic acid) needed by the body to prevent and combat the activities of free radicals. *C. cainito* is a good source of minerals such as potassium, calcium and phosphorus (Table 11), which are needed for electrolyte balance, neurotransmission, and development of strong teeth and bones (Roth and Townsend, 2003).

Results obtained in Table 12 shows that the sample contains protein (1.96 g for pulp and 4.63 g for seed) which indicates high nutritional quality. Crude fibre (4.19 g for seed and 2.31 g for pulp) and ash contents are high and could aid bowel movement and also increase mineral contents respectively in the body (WHO/FAO, 1970). The gross energy values (474.77 for seed and 89.32 for pulp) are high and this could facilitate protein utilization and possibly avert protein-energy malnutrition which is very common in underdeveloped countries evident by high cost of proteins in the diet (WHO/FAO, 1970).

Moisture contents have a great impact in the preservation of food materials. Moisture content is one of the most important and most widely used parameter in food processing. Moisture contents of the star apple pulp and seed are 75.90 and 54.04, respectively. Fat contents of pulp and seed extracts are 0.88 and 15.81g respectively as shown in Table 12.

C. cainito can serve as a good food supplement especially for the obese because of its low fat content. Its low carbohydrate content underscore its low value of simple sugar, hence, it can be consumed by diabetic patients.

Few fruits have the wide range of bioactivity exhibited by *C. cainito* (Luo et al., 2002). It appears to have excellent health and medicinal benefits which deserve to be further explored. Compared with other fruits, *C. cainito* could be classified as one of the new super fruits.

REFERENCES

Adepoju OT, **Adeniji** PO, **2012**. Nutrient composition and micronutrient potential of three widely grown varieties of African Star Apple (*Chrysophyllum albidum*) from Nigeria. Afr J Food Sci, 6(12):344-357.

Arukwe U, Amadi BA, Duru MKC, Agomuo EN, Adindu EA, Odika PC, Lele KC, Egejuru L, Anudike J, **2012**. Chemical composition of *Persea americana* leaf, fruit and seed. IJRRA, 11(2):346-348.

Association of Official Analytical Chemist, **AOAC**, **2000**. Official Methods of Analysis. Seventeenth edition, Association of Official Analytical Chemist International, Gaithersbugh Maryland, USA.

Barnett HI, **Hunters** BB, **1987**. *Illustrated Genera of Imperfect Fungi.* 4th ed. Macmillian Publishing Company, New York, USA. pp 106, 130.

Banso A, **Adeyemo** SO, **2007**. Evaluation of antibacterial properties of tannins isolated from *Dichrostachys cinerea*. Afr J Biotechnol, 6(15):1785-1787.

Beishir l, **1987**. Microbiology in Practice. *A* Self- *Instructions Laboratory Course*, 4th edition, Harper and Row Publishers, New York. pp 96-111, 120-130, 238-272.

Bonire JJ, Jalil NSN, Lori JA, **1990**. Sodium and potassium content of two cultivars of white yam (*Dioscorea rotundata*) and their source soils. J Sci Food Agric, 53:271-274.

Borris RP, **1996**. Natural products research: Perspectives from a major pharmaceutical company. J Ethnopharmacol, 51:29-38.

Buchannan RE, **Gibbon** NE, **2000**. Bergeys Manual of Determinative Bacteriology. Williams and Wilkens Co. Baltimore, USA.

Buhler DR, **2000**. Antioxidant Activities of Flavonoids. Department of Environmental and Molecular Toxicology, Oregon State University, USA.

Chandrasekaran M, **Venkatesalu** V, **2004**. Antibacterial and antifungal activity of *Syzygium jambolonum* seeds. Journal of Ethnopharmacology, 91:105-108.

Chang SK, **2003**. Protein Analysis in Food Analysis. Third edition, Kluwer Academic Plenum Publishers, New York, USA, pp. 134-135.

Cheesbrough M, **2000**. District Laboratory Practice in Tropical Countries, Part 2. Cambridge University press, U.K. pp 35-38, 62-69.

Chung K, Lu Z, Chou MW, **1998**. Mechanism of inhibition of tannic acid and related compounds on the growth of intestinal bacteria. Food Chem Toxicol, 36:1053-1060.

Cowan MM, **1999**. Plant products as antimicrobial agents. Clin Microbiol Rev, 12(4):564- 582.

Dalitha MK, **2008**. Manual on the Antimicrobial Susceptibility Testing. Indian Association of Medical Microbiology. pp 10-21.

Denwick PM, **2002**. Natural Products: A Biosynthetic Approach. 2nd Edn. John Wiley and Sons, Ltd, England. pp 241-243.

Domsch KH, Gams W, Anderson TH, **1993**. Compendium of Soil Fungi. IHW, Eching, Germany.

Duke J, 1992. Handbook of Biological Active Phyto-chemicals and their Activities. CRC press, BICA Ration (FL). pp. 99 – 131.

Einbond LS, Reynertson KA, Luo XD, Basile MJ, Kennelly EJ, **2004**. Anthocyanin antioxidants from edible fruits. J Food Chem, 84(1):23-28.

Erhabor JO, Oshomoh EO, Timothy O, Osazuwa ES, Idu M, **2013**. Antimicrobial activity of the methanol and aqueous leaf extracts of *Emilia coccinea* (Sims) G. Don. Nig J Biotechnol, 25:37-45.

George F, Kerem Z, Makkar HPS, Becker K, **2002**. The biological action of saponins in animal systems: A review. Brit J Nutr, 88(6):587-605.

Harrigan WF, **McCance** ME, **1990**. *Labortory Methods in Food and Diary Microbiology*. 8th Ed., Academic press Inc, London .pp7-23,286-303.

Heslem E, **1989**. Plant Polyphenol: Vegetal Tannin. Telisted-Chemstry and Pharmacology of Natural products. 1st Edn. Cambridge University press, Cambridge, Massachusetts. p169.

Hoge CW, Adams J, Buchanan B, Sears SD, **1991**. Enterococcal bacteremia: To treat or not to treat; a reappraisal. Rev Infect Dis, 13(4):600-605.

James CS, **1995**. Analytical Chemistry of Foods. Chapman and Hall, New York, USA. pp 41-75.

Kirk H, **Sawyer** R, **1998**. Pearson's Composition and Analysis of Foods. Fourth edn, Churchill Livingstone, Edinburgh, Britain. pp 26-28.

Luo XD, Basile MJ, Kennelly EJ, **2002**. Polyphenolic antioxidants from the fruits of *Chrysophyllum cainito* (star apple). J Agric Food Chem, 50(6):1379-1382.

Madigan MT, **Martinko** JM, **2006**. Brock Biology of Microorganisms. 11th Edition. Pearson-Prentice Hall Publishers, Upper Saddle River, NJ, USA. p256.

Nakahara K, **Kawabata** H, **1993**. Inhibitory effects of Oolong tea polyphenols on glucosyltransferases of *Streptococcus mutans*. Appl

Environ Microbiol, 59(4):968-973.
National Research council, NRC, **2008**. Star Apples; Lost Crops of Africa. Vol III : Fruits- Lost crops of Africa. National Academic Press.
Ness AR, Khaw KT, Bingham S, Day NE, **1996**. Vitamin C status and serum lipids. Eur J Clin Nutri, 50:724-729.
Okwu DE, **2001**. Evaluation of the chemical composition of Indigenous spices and flavoring agents. Global J Pure Appl Sci, 7(3):455 -459.
Ologhobo AD, **Fetuga** BI, **1983**. Investigation on the trypsin inhibitor hemagglutinin, phytic and tannic acid contents of cowpea *Vigna unguiculata.* Food Chem J, 12(4):249-254.
Pelczar MJ, Chan ECS, Krieg NR, **1993**. Microbiology Concepts and Applications. 1st Edn. Mchraw-Hill Inc., New York, U.S.A. pp 80-100,158-161, 370.
Roth AR, **Townsend** CE, **2003**. Nutrition and Diet Therapy. 8th Edn. Delmar learning, Thomson learning Inc. Canada. pp 132.
Salah W, Miller NJ, Pangauga T, Bolwell GP, Rice E, Evans C, **1995**. Polyphenolic flavonoids as scavengers of aqueous phase radicals as chain-breaking antioxidant. Arch Biochem Biorh, 2:339– 340.
Sharma K, **2007**. Manual of Microbiology: Tools and Techniques, 2nd Edn., Ane Books Pvt., Limited, New Delhi, India. 405p.
Sodipo OA, **Akiniyi** JA, **2000**. Studies on certain characteristics of extracts from bark of *Pansinystaliama cruceras* (*K. schum*). Pierre Exbeille. Glob J Pure Appl Sci, 6:83–87.
WHO/FAO, **1970**. Reference Protein Manual. World Health Organization/Food and Agricultural Control.
Xu H, **Lee** SF, **2001**. Activity of plant flavonoids against antibiotic-resistant bacteria. Phytother Res, 15:39-43.
Yu Z, Ding X, Xia L, Xiao X, Cao Z, Xu S, Liu X, **2013**. Antimicrobial activity and mechanism of total saponins from Allium Chinese. J Food Sci, 34(15):75-80.

Citation: Oranusi SU, Braide W, Umeze RU, 2015. Antimicrobial activities and chemical compositions of *Chrysophyllum cainito* (star apple) fruit. Microbiol Res Int, 3(3): 41-50.