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Characterisation, Antioxidant Activity and Chain Breaking Properties of *Blighia sapida* Oil Extracts

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Abstract: *The ackee (Blighia sapida), a member of the Sapindaceae family is native to West Africa but has been naturalised in the Caribbean. Fully mature fruits are consumed in Jamaica and its diaspora. The arilli of the mature fruit is rich in monounsaturated lipids. Lipid extracts of the fruit are not currently utilised commercially. In the current study, lipid extracts of the arilli and seeds of the fully mature fruit were characterised. The acid, iodine, peroxide, and saponification values of the lipid extracts, as well as the refractive index were determined. Their antioxidant and chain breaking properties were also evaluated. Ackee arilli oil and seed oil had acid values of 3.2 ± 1.3 mg KOH/g and 4.7 ± 0.3 mg KOH/g, respectively ($p > 0.05$). The refractive index of ackee arilli oil and seed oil was 1.46 and 1.45, respectively ($p > 0.05$). Ackee seed oil had an iodine value of 48 and arilli oil 47 – 56 (g I_2/100 g oil) ($p > 0.05$). Oil extracts from the mature arilli of the fruit had a saponification value of 211-213 mg KOH/g whereas ackee seed oil had a saponification value of 140 mg KOH/g. The peroxide values for the arilli and seed oil samples were 1.88 ± 0.35 and 19.55 ± 2.90 mEq/kg, respectively ($p < 0.05$), indicating that arilli oil had greater stability than ackee seed oil. The reducing capacity of ackee seed oil (3184 ± 526 %) and arilli oil (2078 ± 252 %) was not significantly different ($p > 0.05$). Fractionation of arilli oil extracts revealed that neutral lipids made up the major portion followed by glycolipids. Phospholipids were present in very small quantities. DPPH radical scavenging assay revealed percentage inhibitions of 3.2 % and 20.1 % for arilli and seed oils, respectively. Ackee arilli oil contained higher quantities ($p < 0.05$) of phenolics compounds (14.35 ± 0.66 ppm gallic acid/g) than the seed oil (11.47 ± 0.53 ppm gallic acid/g). Ackee oil exhibited several characteristic features of oils that are currently utilised commercially. Ackee oil extracts could be considered for utilisation in commercial applications in the food and cosmetic industries.*

Keywords: *Blighia sapida, ackee, oil, characterisation, antioxidant*

1. Introduction

Ackee (*Blighia sapida* Koenig), a tropical fruit belonging to the Sapindaceae family is originally from West Africa growing in the region of Senegal to Cameroon and Guinea. It has been naturalised in the Caribbean and in Florida, of the United States of America. The arilli of the fruit is widely consumed in Jamaica but underutilised in West Africa. In Benin, the fruit has emerged as a high priority species for domestication (Ekue et al., 2010).

The fruit consists of a red pod and yellow arilli with black seeds (see Figure 1). Full maturity is attained after seven to eight weeks of development. During week's two to three of development, the fruit doubles in size after which fruit size increases at a slower rate. When fully mature the pods split revealing three to four black shiny seeds surrounded by a yellow aril. Only the arilli of fully mature fruits should be eaten. This is due to the presence of a toxic non-proteinogenic amino acid, hypoglycin A which is present in the unripe fruit (Brown et al., 1992; Blake et al., 2004; Chase et al., 1990).

Figure 1. Fully Mature Ackee Fruit

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Upon metabolism, hypoglycin A is converted to methylenecyclopropaneacetyl coenzyme A which interferes with fat metabolism and leads to a lowering of glucose levels, also known as hypoglycemia (Von Holt, 1966). Several acyl-CoA dehydrogenases which play a role in the Krebs cycle are inhibited (Kean, 1976; Billington et al., 1978). In severe cases coma and death may occur (Hill, 1952). As the fruit matures, hypoglycin A is translocated from the arilli to the seeds of the fruit (Bowen-Forbes et al., 2011). Only then is the fruit safe for consumption. The seeds from mature fruits remain a rich source of hypoglycin A but the concentration within the arilli decreases from over 1000 ppm to undetectable (Brown et al., 1992).

As the fruit matures there is an increase in its lipid content from 0.7 % in the arilli of the immature fruit to over 50 % in the arilli of the fully mature fruit (Goldson, 2002). A decrease in the percentage free fatty acid and acid value of lipid extracts of the arilli was also observed as the fruit matured (Goldson, 2002). At each stage of maturity oleic acid was the predominant fatty acid present followed by palmitic acid and stearic acid (Goldson, 2002). Grande-Tovar et al., (2019) reported a twelve-fold increase in the levels of oleic acid as the fruit matured. The lipid content of the fruit is expected to impact hypoglycin content. Hypoglycin A is an amino acid which is water soluble. As the fruit matures hypoglycin A is converted to hypoglycin B (a dipeptide) which is stored in the seeds of the fruit (Bowen-Forbes and Minott, 2011).

The major antioxidants present in seed oils are tocopherols, phenols and phospholipids (Ramadan et al. 2003). During refining these antioxidants are removed. Edible crude oils therefore exhibit a greater antioxidant effect than their processed counterparts (Ramadan et al., 2003). Antioxidants improve oil stability, sensory and nutritional characteristics (Ramadan et al., 2003).

The ackee is economically important to Jamaica. It is processed as canned ackee in brine and exported primarily to the United States of America, the United Kingdom and Canada. Significant quantities of waste (seeds, rejected arilli, pods) generated during processing could be considered for commercial applications. It is against this background that the physiochemical properties of lipid extracts from the fruit were investigated. The rise in consumer demand for products developed from new vegetable oils which are more environmentally friendly (Prieto Vidal et al., 2018) continue to fuel the need for the development of more agro based products which utilise more renewable raw materials as compared to petrochemical raw materials which are dependent on fluctuating and expensive oil prices.

Several studies have characterised ackee oil from fruits growing in West Africa and Jamaica (Odutuga et al., 1992; Wellington et al., 1999; Goldson, 2002; Emanuel et al., 2013; Goldson et al., 2014; Aladekoyi et al., 2019). The fatty acid profile of the fruit appears to differ based on its geographical location of growth (Odutuga et al., 1992; Wellington; et al., 1999; Goldson, 2002; Emanuel et al., 2013; Goldson et al., 2014; Aladekoyi et al., 2019). In the current study, ackee oil from fruits grown in Jamaica was characterised. The specific objectives of this study were to characterise lipid extracts of the arilli and seed of the mature ackee fruit to determine its suitability for use in commercial applications such as in the surfactant industry or as an edible oil.

2.0 Materials and methods

1 , 1 – Diphenyl – 2 – picrylhydrazyl, Folin – Ciocalteu's Reagent and Wijs solution were supplied by Sigma-Aldrich, St Louis, USA. All solvents were of HPLC grade.

2.1 Drying and milling of ackee fruits

Fully mature ackee fruits were harvested from trees at The University of the West Indies, Kingston, Jamaica. Fruits were separated into their individual components. Arilli and seeds were dried to constant weight (55°C, 6 days, Gallenkamp Laboratory Oven OV-330, England). A slightly lower temperature of 55°C was selected compared to a drying temperature of 60°C utilised by Anderson-Foster et al., (2012) to minimise the loss of antioxidants within the fruit. Dried samples were milled utilising a Perten Instrument, Lab Mill 3600, Springfield, Illinois, USA (30 s, 25°C) and extracted utilising n-hexane (100%, 24 h). The resulting ackee arilli and oil extracts were concentrated *in vacuo* utilising a rotary evaporator (BUCHI Rotavapour R-124).

2.2 Refractive Index

A portable Mettler Toledo digital refractometer (Refractometer 30PX, range of 1.32 - 1.50 nD) was utilised to determine the refractive index of oil extracts.

2.3 Acid Value

The acid value was determined utilising the standard AOAC methods (1990). Ackee oil (1 g) was titrated with neutralised ethanol (10 mL, 100 %) against KOH (0.1 M) using phenolphthalein as indicator. The acid value was calculated according to equation (1).

$$\text{Acid value} = \frac{titer\ (mL\ of\ 0.100\ M)\ x\ 5.61}{Sample\ mass} \qquad (1)$$

2.4 Iodine Value

Oil extracts (0.5-1.0 g) were dissolved in chloroform (100%, 20 mL) and Wijs solution (25 mL, 0.1 mol/L, ICl, Sigma-Aldrich, St Louis, USA) added (American Oil Chemists' Society, 1998). The samples were stored in the dark (1 h) after which KI (30 %, 20 mL) and H_2O (100 mL) were added. The samples were titrated against standardised $Na_2S_2O_3$ (0.1 M). Equation (2) was used to calculate the iodine value (IV).

$$IV = \frac{(b - s) \text{ x N x } 12.69}{\text{Sample mass}} \qquad (2)$$

where:

b = Oil sample

s = Blank titration
N = $Na_2S_2O_3$ normality

2.5 Saponification Value

The saponification value was determined utilising the standard AOAC methods (1990). Oil extracts (4-5 g) were dissolved in standardised alcoholic KOH (0.5 M, 50 mL) and refluxed (2 h). After cooling, samples were titrated with standardised hydrochloric acid (0.5 M). Saponification values (SAP) were obtained according to Equation (3).

$$SAP = \frac{28.05\,(a - b)}{\text{Sample mass}} \quad (3)$$

where:
a = Blank titration
b = Sample titration

2.6 Peroxide Value (PV)

The peroxide value was determined utilising the standard AOAC methods (1990). Oil extracts (1.5 g) were weighed in a conical flask (250 mL). Chloroform (10 mL, 100 %), glacial acetic acid (15 mL, 99.7 %) and freshly prepared saturated aqueous potassium iodide solution (1 mL) were added. The saturated solution was prepared by the addition of excess potassium iodide in freshly boiled water. Observation of solid potassium iodide is evidence of a saturated solution. Flasks were stoppered, shaken (1 min) and placed in the dark (1 min). To each flask was added distilled water (75 mL). The liberated iodine was titrated with sodium thiosulphate (0.01 M) using a starch solution (1 %) as indicator. A blank determination was performed. Equation (4) was used to calculate PV (AOAC 1990).

$$PV = [(V-V_0)\,T)/m] \times 10^3 \text{ mEq/kg} \quad (4)$$

Where:
V = Thiosulphate titer volume
V_0 = Blank volume
T = Molarity of thiosulphate solution
m = Mass of oil

2.7 Reducing Capacity Assay

The reducing capacity of oil extracts was determined as described by Benkeblia (2005). To oil extracts (2 mL) potassium hexacyanoferrate (III), (205 mL, 10 g/L) was added and the mixture incubated (50 – 55°C, 20 min). To this mixture was added trichloroacetic acid (2.5 mL, 100 g/L) and the mixture centrifuged (10 min, 2000 rpm). To the supernatant (2.5 mL) was added distilled water (2.5 mL) and ferric chloride (0.5 mL, 1 g/L). Sample absorbance was measured spectrophotometrically (700 nm, PU8670 VIS/NIR Philips Spectrophotometer). Reducing capacity (RC) was determined from equation (5) (Benkeblia, 2005):

$$RC = [(Am/Ab) - 1] \times 100 \quad (5)$$

where:
Am = Absorbance reaction mixture
Ab = Absorbance blank

2.8 1,1-Diphenyl-2-picrylhydrazyl (DPPH) Assay

DPPH radical scavenging activity was determined based on the protocol from Brand -Williams et al., (1995). Oil extracts (200 µL) were added to DPPH solution (1×10^{-4} M, 2 mL, Sigma-Aldrich, St Louis, USA) which was prepared utilising toluene as solvent. The mixture was centrifuged (10 min, 2000 rpm) and samples incubated in the dark (10 min, 24 °C). The absorbance (515 nm, PU8670 VIS/NIR spectrophotometer, Philips) of the mixture was then determined (Brand-Williams et al., 1995). Toluene was utilised as the blank. Percentage inhibition was calculated according to Equation (6).

$$\%\ \text{Inhibition} = \frac{[A_{DPPH} - A_{SAMPLE}]}{A_{DPPH}} \times 100 \quad (6)$$

where:
A_{DPPH} = Absorbance of DPPH solution
A_{SAMPLE} = Absorbance of sample solution

2.9 Total Phenolic Content

Ackee oil (3 g) was dissolved in n-hexane (6 mL). A methanol-water solution (80:20 v/v, 10 mL) was added and the mixture centrifuged (3000 rpm, 10 min). Hydroalcoholic extracts were removed and the oil phase re extracted with a methanol/water mixture (80:20, v/v) and centrifugation repeated (Ramadan et al., 2003). Hydroalcoholic extracts were combined and concentrated *in vacuo*. The concentrated extracts were dissolved in acetonitrile (15 mL) and residual lipids removed by washing with n-hexane (15 mL x 3). Phenolic extracts were concentrated *in vacuo* and the phenolic content determined as described by (Singleton and Rossi, 1965; Ramadan et al., 2003). Folin-Ciocalteu's reagent (1 mL, Sigma-Aldrich, St Louis, USA) was added to the phenolic extract (0.2 mL) and sodium carbonate (7.5 %, 0.8 mL) added (Singleton and Rossi, 1965). Samples were incubated (30 min) and the absorbance determined (765 nm, PU8670 VIS/NIR spectrophotometer, Philips). A standard curve was generated utilising gallic acid as standard (R^2 = 0.98). For the standard curve, standard solutions of gallic acid were prepared (1 – 20 ppm). 1 mL of each standard solution was utilised in the analysis. The results were expressed as ppm gallic acid/ g sample.

2.10 Chain Breaking Properties

Chain breaking properties of oil extracts were determined based on a procedure adapted from Benkeblia (2005). Oil extracts were heated in a water bath (88 - 92°C, 3 h). To the heated extracts was added DPPH solution (1×10^{-5} M, 3 mL). The mixture was incubated at room temperature (60 min) and the absorbance measured (10 min intervals, 515 nm PU8670 VIS/NIR Spectrophotometer). Chain breaking activity was expressed as the reaction rate based on Equation (7):

$$(1/A^3) - (1/A_0^3) = -3Kt \quad (7)$$

where:
A_0 = Initial optical density
A = Optical density at increasing time, *t*

Ꝁ = Rate constant

A plot of $1/A^3$ vs t produces a slope of -3Ꝁ which was utilised to determine the rate constant.

2.11 Fractionation of Crude Ackee Oil Extracts

Ackee oil extracts were dissolved in chloroform and chromatographed on a glass column (20 mm x 30 cm) packed with an alumina silicate-based ion exchange resin (Hirsch et al., 1972). Neutral lipids were eluted with chloroform, glycolipids with acetone and phospholipids with methanol. Solvents were removed *in vacuo* and the yield determined.

2.12 Statistical Analysis

The mean and the standard deviation of the data were reported. The Student t-test (paired) was conducted to determine any differences between the means at the significant level of $p < 0.05$ (Microsoft Office Excel 2019). Samples were analysed in duplicate except for the oil fractionation which utilised one sample. Table 1 summarises the characteristic properties of ackee arilli and seed oils.

3. Results and Discussion

3.1 Acid Value and Refractive Index

The acid value is an indicator of the level of free fatty acids that are present within an oil. This value can be utilised as an indicator of oil quality or edibility of the oil. A high value implies that there is a high percentage of free fatty acids within the oil which may be due to rancidity. Vegetable oils exist as triglycerides. When rancidity occurs, triglycerides undergo degradation forming glycerol and free fatty acids. Ackee arilli oil and seed oil had acid values of 3.2 ± 1.3 mg KOH/g and 4.7 ± 0.3 mg KOH/g, respectively (see Table 1). These values are not significantly different. An acid value of 1.83 mg KOH/g (Anderson-Foster et al., 2012) and 20.33 mg KOH/g (Wellington et al., 1999) for oil extracts from the mature ackee aril has been previously reported. Sprouted ackee seed oil also has a high acid value of 25 mg KOH/g (Aladekoye et al., 2019).

Different extraction techniques were utilised in the various studies. In the study conducted by Anderson-Foster et al., (2012) arilli were dried in a gravity convection oven and Soxhlet extracted with petroleum ether. Sprouted ackee seed samples were dehulled and sun dried for two weeks prior to extraction (Aladekoyi et al., 2019). It is not clear as to how the seed samples were extracted in the study conducted by Aladekoyi et al., (2019). High acid values indicate the presence of free fatty acids and chemical degradation of the oil due to hydrolytic rancidity. The acid value of ackee arilli oil from the current research is similar to Niger (*Guizotia abyssinica* Cass.) seed oil which has an acid value of 3.1 ± 1.1 mg KOH/g (Negash et al., 2019). The acid values from this research are higher than that typically reported for refined vegetable oil as the samples are unrefined. Acid values of 2.43 ± 0.9 and 0.98 ± 0.23 mg KOH/g oil were reported for local and imported edible oils in Gondar City, Ethiopia (Negash et al., 2019). Further refining of ackee oil extracts would remove any inherent free fatty acids that are present and improve the quality of the oils which could lead to their potential use in the food, pharmaceutical or cosmetic industries.

The refractive index can be utilised to monitor oil quality, detect rancidity and adulteration in edible fats and oils (Arya et al., 1969). Ackee arilli oil (1.46) and seed oil (1.45) had similar values ($p > 0.05$) to olive oil (1.46) (Santonico et al., 2015). Oil obtained from sprouted ackee seeds had a refractive index of 1.46 (Aladekoye et al., 2019). A correlation has been reported between the refractive index and acid value of an oil (Cho et al., 2013). In a study conducted by Xu and Li (2021) the refractive index of peanut oil (1.4631) and Fulinmen colza oil (1.4569) was determined utilising spectrometry. They suggested that the refractive index model and Cauchy matrix of an oil can be utilised as indexes for oil identification (Xu and Li, 2021). The refractive index of ackee oil falls within the range of that reported for other commercial oils again illustrating that it may be considered for use in similar applications.

Table 1. Characterisation and Antioxidant Properties of Ackee Oil

Characteristics	Arilli †	Seed †
Acid value (mg KOH/g)	3.2 ± 1.3a	4.7 ± 0.3a
Refractive index	1.46b	1.45b
Iodine value (g I2/100 g)	47 – 56c	48c
Saponification value (mg KOH/g)	211 – 213d	140e
Peroxide value (mEq/kg)	1.9 ± 0.4f	19.6 ± 2.9g
Reducing capacity (%)	2078 ± 252h	3184 ± 526h
FRSA (%)	3.2i	20.1j
Phenolics (ppm gallic acid/g)	14.4 ± 0.7k	11.5 ± 0.5l
Chain breaking properties	8.67×10^{-2}m	3.56×10^{-3}n
*Oil fractionation g/kg	898.84 (neutral) 94.41 (glycolipids) 21.19 (phospholipids)	946.25 (neutral) 37.46 (glycolipids) 11.40 (phospholipids)

† Values represent the mean ± standard deviation (Number of replicates, N = 2)
Values with the same letter in a row are not significantly different at the 95% confidence level.
* One sample analysed.

3.2 Iodine Value

The iodine value (IV) indicates the level of unsaturation in an oil which directly affects oil stability. Oils with high levels of unsaturation have higher iodine values and are more susceptible to oxidative degradation. Ackee seed oil had an IV of 48 and arilli oil 47 – 56 (g I_2/100 g oil) ($p > 0.05$). This intermediary IV suggests that the oils are monounsaturated. The monounsaturation within arilli oil is due to the presence of oleic acid (Goldson, 2002). Gondoic acid is the main monounsaturated fatty acid present in the seeds of the fruit (Goldson, 2002).

An IV of 49 was also reported for oil from mature ackee arilli (Wellington et al., 1999). Much higher iodine values have been reported for ackee oil from fruits growing in Nigeria with values ranging from 90 – 95 g I_2/100 g oil (mature arilli) and 120 – 132 g I_2/100 g oil (immature arilli) (Adepoju et al., 2013). A lower value of 25 (g I_2/100 g oil) was reported for seed oil extracted from sprouted seeds growing in Nigeria (Aladekoye et al., 2019). Ackees growing in Nigeria have a different fatty acid profile from those grown in Jamaica (Aladekoye et al., 2019). Oleic acid was reported as the major fatty acid in oil extracts from sprouted ackee seeds growing in Nigeria (Aladekoye et al., 2019).

The IV of avocado oil and sunflower oil which also has oleic acid as the predominant fatty acid is 89.5 and 84.6, respectively (Dymińska et al., 2017). These values are higher than that of ackee oil due to the presence of linoleic acid and linolenic acid. Higher percentages of oleic acid are present in sunflower oil (86.2 %). Safflower oil and corn oil which are high in linoleic acid, had iodine values of 161.3 and 129.8, respectively (Dymińska et al., 2017). Saturated fats such as coconut oil have a lower IV ranging from 7.5 – 10 (Gopala et al., 2010). Ackee oil extracts are expected to be more stable to oxidation as compared to safflower oil and corn oil which have higher levels of unsaturation and higher iodine values. Ackee oil is monounsaturated as compared to safflower oil and corn oil which are polyunsaturated.

3.3 Saponification Value

The saponification value (SAP) measures the level of esterification within oils. The SAP value of arilli and seed oils were significantly differently at $p < 0.05$. Oil extracts from the mature arilli had a higher SAP value of 211 – 213 mg KOH/g compared to the seed (140 mg KOH/g). These values are slightly higher than those reported by Adepoju et al., (2013) for mature fruits, (190 - 201 mg KOH/g) and immature fruits, (176 – 198 mg KOH/g). A SAP value of 197 mg KOH/g (Anderson-Foster et al., 2011) and 194 mg KOH/g (Wellington et al., 1999) have also been reported for oil extracts from mature ackee arilli growing in Jamaica. Sprouted ackee seed oil had a SAP value of 225 mg KOH/g (Aladekoye et al., 2019). Popular oils utilised in soap making include castor oil, corn oil and coconut oil which have saponification values of 180, 192 and 268 respectively. This implied that the ackee arilli oil could be considered for utilisation in soap production as its SAP value falls within the range of oils traditionally utilised to make soaps.

3.4 Peroxide Value

Peroxides are formed during oil oxidation and are indicative of rancidity. The peroxide values (PV) for the arilli and seed oil samples were 1.88 ± 0.35 and 19.55 ± 2.90 mEq/kg respectively ($p < 0.05$), indicating that arilli oil had greater stability than ackee seed oil which may be more susceptible to oxidation. Ackee arilli oil is rich in oleic acid and low in polyunsaturated fatty acids enhancing its stability (Goldson 2002). Ackee seed oil has a higher level of monounsaturation and is rich in both gondoic acid and oleic acid (Goldson-Barnaby and Williams, 2017). Oils with higher levels of unsaturation are more susceptible to oxidation. Adepoju et al. (2013) reported PV values of less than 10 mEq/kg for the immature and mature arilli of the fruit. Commercial vegetable oils have a PV of less than 10 mEq/kg. PV values of 1.46, 6.32 and 6.53 have been reported for sunflower, avocado, safflower oils (Dymińska et al., 2017). Oils having PV values between 30 – 40 mEq/kg exhibit a rancid taste (Godswill et al., 2018).

3.5 Reducing Capacity Assay and Lipid Fractionation

In the reducing capacity assay, the ability of a sample to reduce Fe^{3+} to Fe^{2+} was evaluated (Elmastas et al., 2006). The formation of Fe^{2+} produced a green solution, the intensity of which was proportional to the quantity of Fe^{2+} formed. The higher the absorbance, the greater the reducing power of the sample. The reducing capacity of a sample may be used as an indicator of its potential antioxidant activity (Elmastas et al., 2006; Gulcin et al., 2004). The reducing capacity of ackee seed oil (3184 ± 526 %) and arilli oil (2078 ± 252 %) was not significantly different ($p > 0.05$). Ackee seed and arilli oils exhibited a higher reducing capacity than those from aqueous extracts of onion varieties (20 – 100 %) and garlic (approximately 200 %) as reported by Benkeblia (2005). This suggests that the oils possess antioxidant properties which may be due to the presence of phenolics within the oil extracts.

Fractionation of ackee arilli and seed oil extracts revealed that neutral lipids made up the major portion of the oil followed by glycolipids with phospholipids being present in very small amounts (see Table 1). Glycolipids and phospholipids are expected to be more soluble in the aqueous reaction medium than neutral lipids. Both arilli and seed oils had a high reducing capacity thus indicating that the oils exhibit antioxidant properties.

3.6 Antioxidant Properties and Free Radical Scavenging Activity

The free radical scavenging activity (FRSA) of ackee lipid extracts was investigated utilising the DPPH assay. As the DPPH radical is reduced the colour intensity of the solution decreases. The percentage inhibition of arilli and

seed oil was 3.2 % and 20.1 %, respectively (see Table 1). No further change in percentage inhibition was observed when the length of incubation period was further increased. Ackee seed oil exhibited a significantly higher ($p < 0.05$) FRSA activity than that of arilli oil. This suggests the presence of other components within the oil, such as phenolics which are antioxidants. The antioxidant properties of aqueous extracts of the ackee fruit and arilli oil have been reported (Dossou et al., 2014; Goldson-Barnaby et al., 2018). Grande-Tovar et al. (2019) investigated the bioactive micro-constituents within ackee arilli and found that as the fruit ripened there was an increase in antioxidant activity. Whereas there was a decline in the levels of ascorbic acid and polyphenolics, there was an increase in the levels of oleic acid, squalene, and D:A-Friedooleanan-7-ol, (7.alpha.) which positively impacted the antioxidant capacity of the fruit (Grande-Tovar et al., 2019). Squalene, a triterpene, would significantly contribute to the antioxidant activity of the fruit (Grande-Tovar et al., 2019). Ackee seed oil contained several bioactive constituents inclusive of tannins, saponins, phenols, alkaloids and flavonoids which contributed to its free radical scavenging and antioxidant properties (Onuekwusi et al., 2014).

3.7 Total Phenolic Content

Ackee arilli oil contained significantly higher quantities ($p < 0.05$) of phenolic compounds (14.35 ± 0.66 ppm gallic acid/g) as compared to the seed oil (11.47 ± 0.53 ppm gallic acid/g) (see Table 1). It should be noted that several studies have shown that there is no direct correlation between the amount of phenolics in a sample and its antioxidant activity (Gulcin et al., 2004; Ramadan et al., 2003). Other bioactive compounds within the fruit contributes to its antioxidant properties (Dossou et al., 2014; Grande-Tovar et al., 2019). As the fruit ripened the level of phenolics increased in the seed and husk of the fruit while decreasing in the arilli (Sybron et al., 2019).

3.8 Chain Breaking Properties

The chain breaking activity for arilli oil (unheated, 8.70 x 10^{-2} $OD^{-3}min^{-1}ml^{-1}$; heated, 2.52 x 10^{-1} OD^{-3} $min^{-1}ml^{-1}$) and seed oil (unheated, 3.56 x 10^{-3} $OD^{-3}min^{-1}ml^{-1}$; heated, 3.76 x 10^{-2} OD^{-3} $min^{-1}ml^{-1}$) samples increased after heating. An increase in chain breaking activity is accompanied by a decrease in the reducing capacity of the samples and a decrease in total phenolics (Benkeblia et al., 2005). Polyunsaturated fatty acids such as linoleic acid and alpha-linolenic acid exhibit antioxidant properties protecting the body from oxidative stress (Richard et al., 2008). Chain breaking results in shorter fatty acid chains and a reduction in reducing power. Non-enzymatic browning causes the polymerisation of phenolic compounds forming brown coloured macromolecules (Benkeblia, 2005). This suggests that heating reduces antioxidant activity. The chain breaking properties (OD^{-3} $min^{-1}ml^{-1}$) of the ackee lipid extracts significantly increased as the samples were heated (arilli oil: 8.67x10^{-2} (unheated) to 2.52 x 10^{-1}(heated); seed oil: 3.56 x 10^{-3} (unheated) to 3.76 x 10^{-3}(heated).

4. Conclusion

Ackee arilli and seed oil extracts are currently not utilised commercially. They however show promising characteristic properties for utilisation as an edible oil or in the cosmetic industry. The oils had similar characteristics to that of other commercially available oils. Ackee arilli oil is expected to be stable to oxidation due to its low levels of unsaturation which was confirmed from its iodine value. Its saponification value illustrates its potential application in the surfactant industry. The antioxidant properties of the oils are shown from their free radical scavenging activity and phenolic content. The peroxide value revealed that ackee seed oil was more oxidised than arilli oil. Ackee lipid extracts may be utilised in the production of value-added food commodities as well as in the cosmetics industry. The saponification value, iodine value, and fatty acid composition of ackee oil exhibit similarity to that of other oils which are utilised commercially.

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