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Phytochemical Composition and Radical Scavenging Activity of Hexane, Ethyl acetate and Methanol Extracts of *Spermacoce ocymoides* (Burm F.) DC and *Annona muricata* L. Leaves and Fruit Juice

[1*]Salau, A. K., [1]Omolola, O. S., [1]Raji, H. O., [2]Ajiboye, T. O., [3]Ogunbode, S. M., [4]Yakubu, M. T. and [5]Oladipo, O. T.

[1]Antioxidants, Toxicology and Phytomedicine Research Laboratory, Biochemistry and Nutrition Unit, Department of Chemical Sciences, Fountain University, Osogbo, Nigeria.
[2]Antioxidants, Free Radicals, Functional Foods and Toxicology Research Laboratory, Department of Biological Sciences, Al-Hikmah University, Ilorin, Nigeria.
[3]Nutritional Biochemistry Research Laboratory, Biochemistry and Nutrition Unit, Department of Chemical Sciences, Fountain University, Osogbo, Nigeria.
[4]Phytomedicine, Toxicology and Reproductive Biochemistry Research Laboratory, Department of Biochemistry, University of Ilorin, Nigeria.
[5]Department of Botany, Obafemi Awolowo University, Ile-Ife, Nigeria.

Abstract: This study investigated the *in vitro* free radical scavenging capacity and phytochemical constituents of methanol, ethyl acetate and n-hexane extracts of leaves of *Spermacoce ocymoides* (Burm F.) DC (Rubiaceae) and *Annona muricata* L. (Annonaceae) as well as the fruit juice of *A. muricata*. *A. muricata* fruit juice was also investigated for its vitamin composition. The radical scavenging activities of the solvent extracts of the leaves and fruit juice were determined by measuring their reducing activities against 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical, hydrogen peroxide and ferric ion. The methanol, ethyl acetate and hexane extracts of *S. ocymoides* and *A. muricata* leaves contained phenolics, alkaloids and tannins, while only the methanol and ethyl acetate extracts of both plants contained flavonoids and triterpenes, respectively. Saponins were present in the hexane extracts of both plants and ethyl acetate extract of *A. muricata* leaves. Steroids, phlobatannins and glycosides were not detected in the extracts. *A. muricata* fruit juice contained the highest amount of flavonoids while the *A muricata* methanol leaf extract contained the least amount of flavonoids. *S. ocymoides* methanol leaf extract contained the highest amount of phenolics, while the *A muricata* ethyl acetate leaf extract contained the least amount of phenolics. *A muricata* fruit juice contained more vitamin C than vitamin B_1, with vitamin B_2 having the lowest concentration. The extracts of the leaves of both plants as well as *A. muricata* fruit juice reduced DPPH, hydrogen peroxide and ferric ion at concentrations varying from 0.2 mg/ml to 1.0 mg/ml. Overall, the results showed that the extracts of *S. ocymoides* and *A. muricata* leaves as well as *A. muricata* fruit juice contained antioxidants and vitamins and also exhibited *in vitro* free-radical scavenging properties, which might have been responsible for the acclaimed and the reported properties of these plants.

KEYWORDS: *Spermacoce ocymoides*, *Annona muricata*, vitamins, antioxidants, free radicals

1.0 Introduction

Antioxidants are known to scavenge endogenous free radicals that are thought to cause oxidative stress, which may lead to degenerative diseases such as cancer. Several medicinal plants have been screened for their antioxidant properties, while others are yet to be screened. Those claimed to possess antioxidant properties with little or no scientific validation are *Spermacoce ocymoides* and *Annona muricata* leaves and fruit juice.

Spermacoce ocymoides (Burm F.) DC (Rubiaceae), also known as Purple leaved button weed and Basil like button weed (English) as well as Irawo-ile (Yoruba, southwest Nigeria), is a vascular plant without significant woody tissue above or at the ground level. It is found in India,

***Corresponding Author**
Tel.: +2348064810736
E-mail: ka_salaudeen@yahoo.com

Cameroon, Congo, Gabon, Uganda and as a wayside weed in Osogbo (southwest Nigeria). There are folkloric and ethno-medicinal claims that *S. ocymoides* can be used in the treatment of bacterial infections. The plant was also reported to have wound healing properties, used to treat diarrhoea and dysentery, and also used in the treatment of ring worm, eczema and other skin diseases (Ayyanar and Ignacimuthu, 2009; Shanmugam *et al*., 2011; Conserva and Ferreira Jr., 2012; Onawumi *et al*., 2012). It is also reported to contain saponins, volatile oils and also possess antibacterial activity (Onawumi *et al*., 2012). *S. ocymoides* roots have also been reported to contain steroids, reducing sugars, phenolics and tannins (Jeyachandran *et al*., 2013).

Annona muricata L. (Annonaceae), locally called sharp sharp or soursop in Nigeria and Graviola in south America, is a broad-leaved, flowering, evergreen tree native to tropical America, but now widespread in the tropics. It is adapted to areas of high humidity and relatively warm winters attaining a height of about 8 m (Burkill, 1985). *A. muricata* is abundant in southern Nigeria where it is known for its succulent white endocarp. The fruit pulp is soft with an agreeably sour flavour. It is usually eaten raw and contains fibre. The flavour has been described as a combination of strawberry and pineapple, with sour citrus flavour notes contrasting with an underlying creamy flavour reminiscent of coconut or banana. The fruit contains significant amounts of vitamins B_1, B_2 and C (Morton, 1987). There is evidence indicating that the fruit extracts selectively inhibit the growth of human breast cancer cells by down regulating expression of epidermal growth factor receptor (EGFR) *in vitro* and in a mouse model (Dai *et al*., 2011). The ethanolic leaf extract has been reported to contain enzymic and non-enzymic antioxidants such as phenolics, flavonoids and vitamin C as well as to also scavenge free radicals such as hydrogen peroxide and DPPH (Baskar *et al*., 2007; Muthu and Durairaj, 2015). The seeds, bark and pulp have also been reported to possess cytotoxic activity against human cell lines as well as antioxidant and antimicrobial activities (Ahalya *et al*., 2013; Vijayameena *et al*., 2013; Boakye *et al*., 2015; Raybaudi-Massilia *et al*., 2015).

While studies reporting the antioxidant activities of *S. ocymoides* remain scanty, there has not been any report comparing the phytochemical composition of hexane, ethyl acetate, and methanolic extracts of these two plants with *A. muricata* fruit.

This study therefore compares the phenolic and flavonoid composition of hexane, ethyl acetate, and methanolic leaf extracts of *S. ocymoides* and *A. muricata* and also investigates their phytochemical composition and *in vitro* free-radical scavenging activity, in addition to the vitamin composition of *A. muricata* fruit juice, which is an underutilized fruit tree.

2.0 Materials and Methods

2.1 Plant Materials

S. ocymoides leaves were collected from the premises of Fountain University, Osogbo, Nigeria, while fruits and leaves of *A. muricata* were obtained from a compound in Otta, Ogun State, Nigeria. Both *S. ocymoides* and *A. muricata* were authenticated at the IFE Herbarium, Ile-Ife, Nigeria with voucher numbers 16958 and 17243 respectively.

2.2 Chemicals and Reagents

DPPH, 2, 4-dinitrophenyl hydrazine (DNPH), Folin Ciocalteu's phenol reagent, quercetin and hydrogen peroxide were products of Sigma Aldrich, St-Louis USA. Methanol, ethyl acetate and n-hexane were obtained from Scharlab S.L., Gato Perez, 33-P. I. Mas d'En Cisa. Other chemicals and reagents used were of analytical grade and prepared in glass distilled water.

2.3 Extraction Procedures

Leaves of both plants were air-dried and blended. The resulting powdered sample (70 g for *S. ocymoides*) was successively extracted in 300 ml each of n-hexane, ethyl acetate and methanol with continuous shaking using orbital shaker maintained at 300 rpm for 24 hours each. *A. muricata* sample (70 g) was treated similarly with 550 ml of n-hexane and 300 ml each of ethyl acetate and methanol. The resulting extracts were separately filtered using Whatman

No. 1 filter paper and the filtrates concentrated on water bath to give n-hexane (0.68 g), ethyl acetate (1.53 g) and methanol (4.64 g) extracts for *S. ocymoides* and 4.08 g, 6.47 g and 2.84 g concentrates for the n-hexane, ethyl acetate and methanol, respectively for *A. muricata* leaves.

A. muricata fruit juice was obtained by blending 100 g of the fresh fruit pulp with 100 ml of distilled water after all its seeds have been removed. The blended pulp was then filtered to obtain the juice.

2.4 Phytochemical Screening

The leaf extracts of both plants were screened for their secondary metabolite constituents according to the standard methods of Harbone (1973) and Odebiyi and Sofowora (1978) as follows:

2.4.1 Determination of Secondary Metabolites

2.4.1.1 Alkaloids

To 1 ml of 1% HCl, 3 ml of the extract was added and heated for 20 minutes, cooled, and filtered. A few drops of Wagner's reagent (2 g of iodine and 6 g of KI in 100 ml of distilled water) were then added to 1 ml of the filtrate. A reddish brown precipitate indicated the presence of alkaloids.

2.4.1.2 Tannins

To 1 ml of freshly prepared 10% KOH, 1 ml of the extract was added. A dirty white precipitate indicated the presence of tannins.

2.4.1.3 Phenolics

To 2 drops of 5% $FeCl_3$, 1 ml of the extract was added. A greenish precipitate indicated the presence of phenolics.

2.4.1.4 Glycosides

To 10 ml of 50% H_2SO_4, 1 ml of the extract was added, heated in boiling water for 15 minutes, and 10 ml of Fehling's solution was added and boiled again. A brick-red precipitate indicated the presence of glycosides.

2.4.1.5 Saponins

To 5 drops of olive oil, 3 ml of the extract was added in a test tube and shaken vigorously. A stable emulsion indicated the presence of saponins.

2.4.1.6 Flavonoids

To 1 ml of 10% NaOH, 3 ml of the extract was added. A yellow coloration indicated the presence of flavonoids.

2.4.1.7 Steroids

To 5 drops of concentrated H_2SO_4, 1 ml of the extract was added. A red coloration indicated the presence of steroids.

2.4.1.8 Phlobatannins

A few drops of 1% HCl were added to 3 ml of the extract. A red precipitate indicated the presence of phlobatannins.

2.4.1.9 Triterpenes

To 1 ml of the extract, 5 drops of acetic anhydride was added, followed by a drop of concentrated H_2SO_4. The mixture was steamed for 1 hour and neutralized with NaOH, followed by chloroform. A blue-green colour indicated the presence of triterpenes.

2.4.2 Quantitative Determination of Total Phenolics, Total Flavonoids and Vitamins B and C

Secondary metabolites and vitamins were quantitatively determined using previously described methods:

2.4.2.1 Total Phenolics

This was determined using Folin-Ciocalteu's method, as described by Olajire and Azeez (2011). The extract (0.5 ml) was added to 10 ml deionized distilled water and 2.5 ml of 0.2 N Folin-Ciocalteu's phenol reagent. The mixture was left undisturbed at room temperature for 5 minutes and then 2 ml of 2% sodium carbonate

was added. The absorbance of the resulting solution was read at 780 nm and repeated three times. Quercetin was used as a standard for calibration curve. This was done in triplicate.

2.4.2.2 Total Flavonoids

This was assayed using the procedure described by Jagadish *et al*. (2009). Briefly, the extract (1.5 ml) was added to 1.5 ml of 2% methanolic $AlCl_3$ solution. The mixture was vigorously shaken on orbital shaker for 5 minutes at 200 rpm and the absorbance was read at 367 nm after 10 minutes of incubation. Quercetin was used as a standard for the calibration curve. The assay was carried out in triplicate.

2.4.2.3 Thiamine (Vitamin B_1)

This was assayed as described by Harisaranraj *et al*. (2009). Briefly, 5 g of the sample were homogenized with ethanolic sodium hydroxide (50 ml). It was filtered into a 100 ml flask and 10 ml of the filtrate was pipette. The colour developed by addition of 10 ml of potassium dichromate and the absorbance was read at 360 nm. A blank sample was prepared and treated as the sample. The assay was carried out in triplicate.

2.4.2.4 Riboflavin (Vitamin B_2)

This was also assayed as described by Harisaranraj *et al*. (2009). A known amount (5 g) of the sample was extracted with 100 ml of 50% ethanol solution and shaken for 1 hour. This was filtered into a 100 ml flask; 10 ml of the extract was pipetted into 50 ml volumetric flask followed by addition of 10 ml of 5% potassium permanganate and 10 ml of 30% H_2O_2 and allowed to stand over a hot water bath for about 30 minutes. Thereafter, 2 ml of 40% sodium sulphate was added. This was made up to 50 ml mark and the absorbance was read at 510 nm. The assay was carried out in triplicate.

2.4.2.5 Ascorbic Acid (Vitamin C)

This was determined as described by Omaye *et al*. (1979). The extract (0.5 ml) was mixed thoroughly with 1.5 ml of 6% TCA and centrifuged for 10 minutes at 3500 *g*. After centrifugation, 0.5 ml of the supernatant was mixed with 0.5 ml of DNPH reagent and allowed to stand at room temperature for an additional 3 hours then added 2.5 ml of 85% sulphuric acid and left undisturbed for 30 minutes. Then the absorbance was read at 530 nm. A set of standards containing 10-50 µg of ascorbic acid were taken and processed similarly along with a blank. The assay was carried out in triplicate.

2.4.3 Determination of in vitro Free Radical Scavenging Activity

The *in vitro* free radical scavenging activity of the extracts was determined as previously described:

2.4.3.1 DPPH Radical Scavenging Capacity

The ability of the extracts to bleach the purple colour of DPPH radical was determined as described by Turkoglu *et al*. (2007). Briefly, 2 ml of various concentrations (0.2–1.0 mg/ml) of each extract was added separately to 2 ml of 0.1 mmol/L methanolic solution of DPPH. After 30 minutes of incubation in the dark at room temperature, the absorbance was read against a control at 517 nm. The scavenging rate (I%) on the DPPH radical was calculated using the expression:

$$I\% = [(A_{control} - A_{sample}) / A\ control)] \times 100$$

Where $A_{control}$ is the absorbance of the control reaction (containing all reagents except the test compound) and A_{sample} is the absorbance of the test compound. The procedure was carried out in triplicate.

2.4.3.2 Hydrogen Peroxide Scavenging Activity

The ability of the extracts to reduce hydrogen peroxide to water and oxygen was determined according to the procedure described by Ruch *et al*. (1989). Briefly, 4 mM of hydrogen peroxide was prepared in phosphate buffered saline (pH 7.4). A known volume (4 ml) of various concentrations (0.2–1.0 mg/ml) of each extract was added to 0.6 ml of hydrogen peroxide

solution. The absorbance was read 10 minutes later at 230 nm against a blank solution containing sample without hydrogen peroxide. The inhibition rate (I%) on the hydrogen peroxide was calculated using the expression:

$$I\% = [(A_{control} - A_{sample}) / A_{control})] \times 100$$

Where $A_{control}$ is the absorbance of the control reaction (containing all reagents except the test compound) and A_{sample} is the absorbance of the test compound. The procedure was carried out in triplicate.

2.4.3.3 Ferric ion Reducing Ability

The ability of the extracts to reduce Fe^{3+} to Fe^{2+} was evaluated by adopting the procedure described by Oyaizu (1986). Briefly, various concentrations of the extracts (0.2-1.0 mg/ml) were suspended in 1 ml of distilled water, mixed with 250 µl of 0.2 M phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide [$K_3Fe(CN)_6$]. The mixture was incubated at 50 °C for 20 minutes, and then 250 µl of 10% trichloroacetic acid was added. Following centrifugation at 604 x *g* for 10 minutes, 250 µl of the supernatant was mixed with an equal amount of distilled water and 0.5 ml of 0.1% $FeCl_3$. The absorbance of the resulting solution was read at 700 nm. The procedure was carried out in triplicate.

2.5 Statistical Analysis

Data were expressed as mean ± standard deviation and significant differences were determined by Analysis of Variance (ANOVA) and Tukey's post hoc test for multiple comparisons at 95% confidence level using SPSS soft-ware (SPSS Inc., Chicago, IL, USA).

3.0 Results

The methanol, ethyl acetate and hexane extracts of *S. ocymoides* and *A. muricata* leaves contained phenolics, alkaloids and tannins, while only the methanol and ethyl acetate extracts of both plants contained flavonoids and triterpenes, respectively. Saponins were present in the hexane extracts of both plants and ethyl acetate extract of *A. muricata* leaves. However, steroids, phlobatannins and glycosides were not detected in any of the extracts (Table 1).

A. muricata fruit juice contained more flavonoids than the methanol extract of *S. ocymoides* leaves while the *A muricata* methanol extract contained the least amount of flavonoids (Table 2).

S. ocymoides methanol leaf extract contained more phenolics than the *A muricata* fruit juice, while the *A muricata* ethyl acetate leaf extract contained the least amount of phenolics (Table 2). Only the *A muricata* fruit juice was investigated for vitamins, which contained more vitamin C than vitamin B_1, with vitamin B_2 having the lowest concentration (Table 2).

The methanol extracts of *S. ocymoides* leaves produced higher inhibition activity against DPPH radical than the ethyl acetate extract with the hexane extract producing the least inhibition against the radicals (Figure 1). The ethyl acetate and hexane extracts of *S. ocymoides* leaves produced higher reducing activity against hydrogen peroxide than the methanol extract (Figure 2), while the methanol extract produced higher reducing activity than the ethyl acetate and hexane extracts (Figure 3).

The *A. muricata* fruit juice produced the highest inhibition against DPPH radical followed by the methanol and hexane leaf extracts. The ethyl acetate leaf extract produced the lowest activity against DPPH (Figure 4).

The fruit juice also produced the highest reducing activity against hydrogen peroxide at 0.2 – 1.0 mg/ml, while the ethyl acetate leaf extract produced the lowest activity at 0.2 and 0.4 mg/ml (Figure 5).

While the fruit juice produced the lowest reducing activity against ferric ions at 0.2 mg/ml, the ethyl acetate leaf extract produced the lowest activity at 1.0 mg/ml with the hexane leaf extract producing the highest activity at this concentration (Figure 6).

4.0 Discussion

This study investigated the phytochemical composition of hexane, ethyl acetate and methanol leaf extracts of *S. ocymoides* and *A. muricata* as well as the vitamin composition of juice from *A. muricata* fruit pulp. The extracts and fruit juice were also investigated for their

Table 1: Phytochemical composition of hexane, ethyl acetate and methanol extracts of *S. ocymoides* and *A. muricata* leaves

Constituent	*S. ocymoides* leaves			*A. muricata* leaves		
	n-Hexane	Ethyl acetate	Methanol	n-Hexane	Ethyl acetate	Methanol
Flavonoids	-	-	+	-	-	+
Phenolics	+	+	+	+	+	+
Alkaloids	+	+	+	+	+	+
Steroids	-	-	-	-	-	-
Phlobatannins	-	-	-	-	-	-
Saponins	+	-	-	+	+	-
Tannins	+	+	+	+	+	+
Triterpenes	-	+	-	-	+	-
Glycosides	-	-	-	-	-	-

S. ocymoides = *Spermacoce ocymoides*, *A. muricata* = *Annona muricata*, + = detected;
- = not detected

Table 2: Flavonoids, phenolics and vitamins in hexane, ethyl acetate and methanol extracts of *S. ocymoides* and *A. muricata* leaves and fruit juice

Const.	*S. ocymoides* leaves			*A. muricata*			
				Leaves			Fruit juice
	n-Hexane	Ethyl acetate	Methanol	n-Hexane	Ethyl acetate	Methanol	
FLA	-	-	0.012 ± 0.001^{b}	-	-	0.002 ± 0.000^{a}	0.026 ± 0.001^{c}
PHE	0.087 ± 0.000^{f}	0.013 ± 0.000^{e}	0.094 ± 0.000^{g}	0.003 ± 0.001^{c}	0.001 ± 0.000^{a}	0.002 ± 0.000^{b}	0.009 ± 0.002^{d}
Vit. B_1	ND	ND	ND	ND	ND	ND	29.440±4.860
Vit. B_2	ND	ND	ND	ND	ND	ND	0.171±0.002
Vit. C	ND	ND	ND	ND	ND	ND	167.740±17.700

Const. = constituent, *S. ocymoides* = *Spermacoce ocymoides*, *A. muricata* = *Annona muricata*, FLA = Flavonoids (mg/ml quercetin), PHE = phenolics (mg/ml quercetin), Vit. B_1 = vitamin B_1 (mg/dl), Vit. B_2 = vitamin B_2 (mg/dl), Vit. C = vitamin C (mg/dl), - = not detected, ND = not determined.

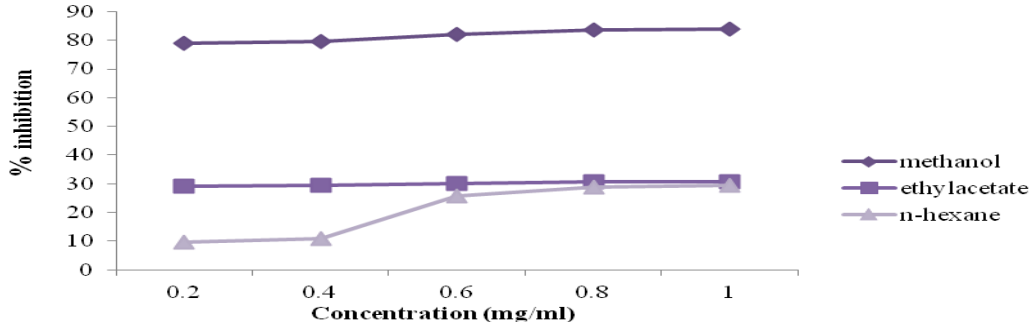

Figure 1: DPPH scavenging activity of methanol, ethyl acetate and n-hexane extracts of *Spermacoce ocymoides* leaves

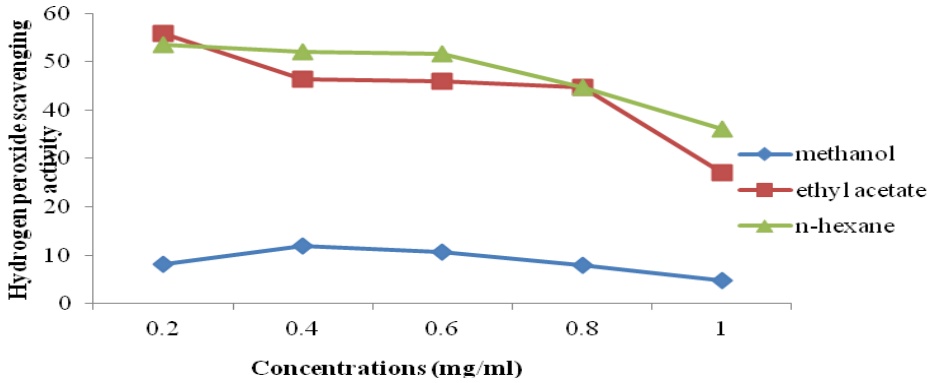

Figure 2: Hydrogen peroxide decomposition activity of methanol, ethyl acetate and n-hexane extracts of *Spermacoce ocymoides* leaves

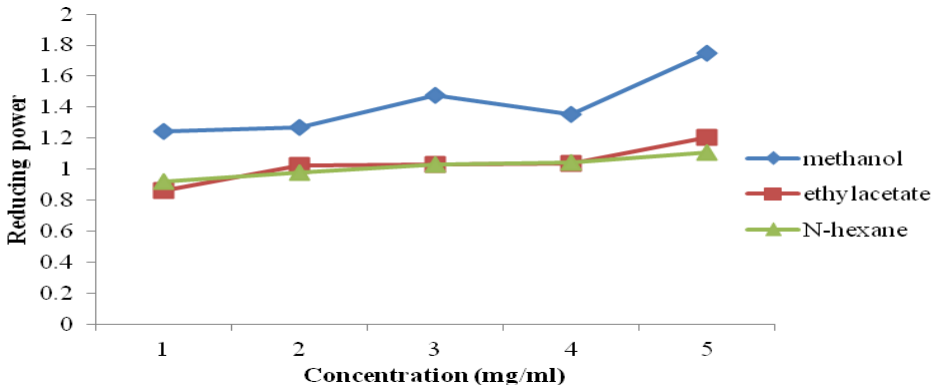

Figure 3: Ferric ion reducing ability of methanol, ethyl acetate and n-hexane extracts of *Spermacoce ocymoides* leaves

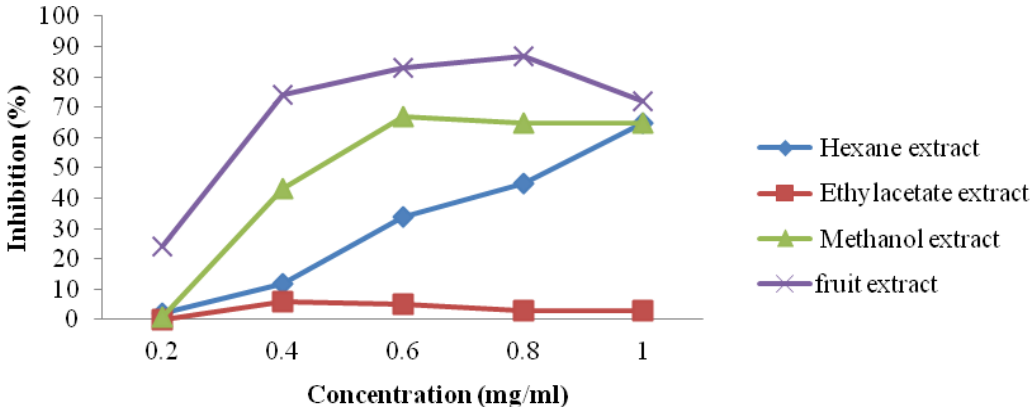

Figure 4: DPPH radical scavenging activity of fruit juice and methanol, ethyl acetate and n-hexane extracts of *A. muricata* leaves

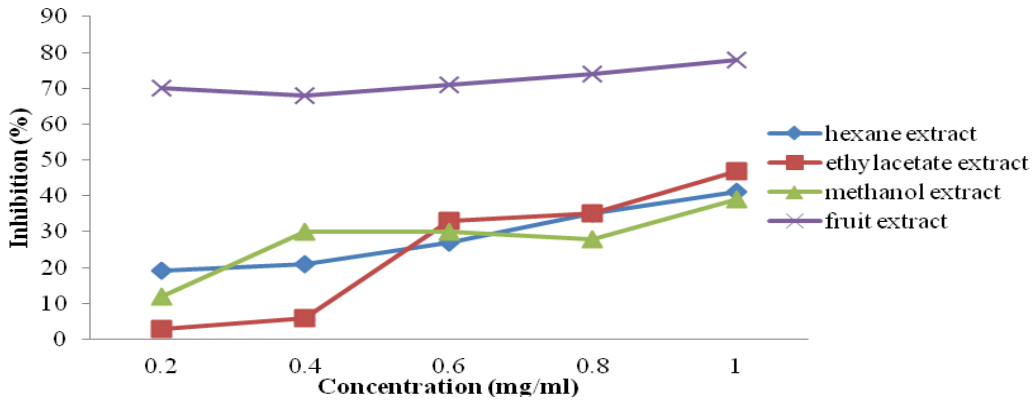

Figure 5: Hydrogen peroxide decomposition activity of fruit juice and methanol, ethyl acetate and n-hexane extracts of *A. muricata* leaves

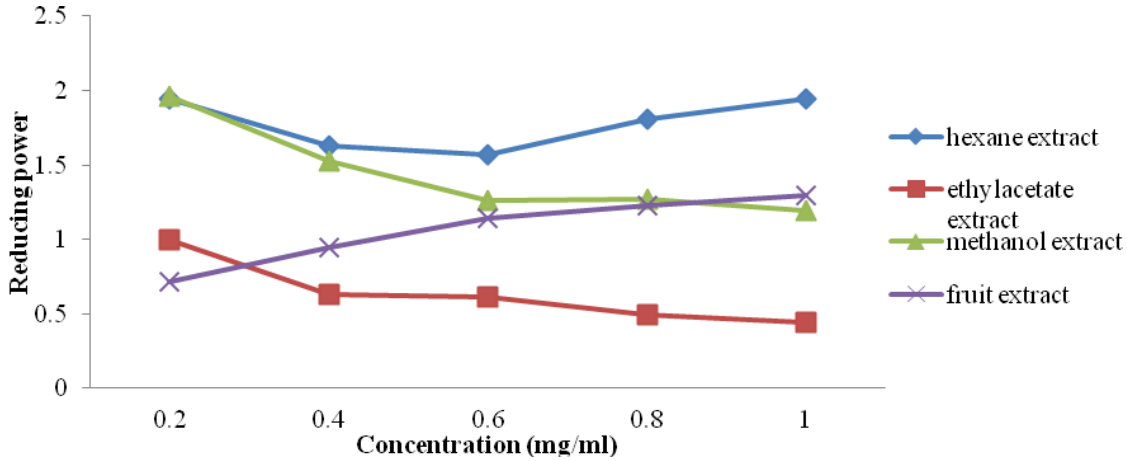

Figure 6: Ferric ion reducing activity of fruit juice and methanol, ethyl acetate and n-hexane extracts of *A. muricata* leaves

ability to reduce DPPH radical, hydrogen peroxide and ferric ions.

Plants have been used over the years not only for their nutritional value but for their therapeutic potential. The presence of secondary metabolite such as flavonoids, phenolics, saponins e.t.c. in plant products or extract influences their biological effects.

The presence of flavonoids, phenolics, alkaloids, saponins, tannins and triterpenes in the hexane, ethyl acetate and methanol extracts of *S. ocymoides* and *A. muricata* leaves would confer a host of pharmacological abilities on these plant extracts. Flavonoids have been reported to possess free radical scavenging and anticancer activities (Nafiu *et al*., 2011; Okigbo *et al*., 2009; Oskoueian *et al*., 2011). Phenolics have also been reported to exhibit tumor inhibiting activity and to inactivate carcinogens and mutagens (Urquiaga and Leighton, 2000; Okwu and Okwu, 2004). Alkaloids are anaesthetics, stimulants and anticancer agents (Upadhyay, 2011) and saponins are cytotoxic and cholesterol-lowering agents (Okigbo *et al*., 2009). Saponins also possess antimicrobial, insecticidal, molluscicidal, antiviral, antifungal, membrane-permeabilising, immunostimulant, hypocholesterolaemic and anticarcinogenic properties (Yücekutlu and Bildacı, 2008). Tannins exhibit antioxidant and antimicrobial properties; they also inhibit viral reverse transcriptase activity and possess protein-

precipitating and iron-binding activities (Shahjahan *et al.,* 2004; Shahjahan *et al.,* 2005; Upadhyay, 2011). Triterpenes have been reported to possess antimicrobial activity (Moodely *et al*., 2011). Thiamine and riboflavin act as coenzymes in dehydrogenase enzymes and take part in redox reactions, while vitamin C acts as an antioxidant and oxygen radical scavenger. They also prevent vitamin deficiency diseases (Bender and Mayes, 2003). These chemical compounds might have been responsible for the acclaimed and reported properties exhibited by the leaves and fruit of these plants such as wound healing, antibacterial, antifungal, radical scavenging, antioxidant and cytotoxic properties (Olajire and Azeez, 2011).

The ability of extracts to reduce DPPH radical has been widely used to assess the *in vitro* free radical scavenging abilities of extracts (Ghasemzadeh *et al*., 2010; Oskoueian *et al*., 2011; Salau *et al*., 2015). Antioxidants scavenge DPPH radical by donating a phenolic hydrogen atom or an electron to it (Olajire and Azeez, 2011). Reduction of hydrogen peroxide has also been used to assess the *in vitro* antioxidant capabilities of extracts (Oloyede *et al*., 2012; Salau *et al*., 2015). Flavonoids have been reported to scavenge hydrogen peroxide by donating electrons to it thereby forming water. The ability of extracts to reduce Fe^{3+} to Fe^{2+} in the ferric ion assay has also been used to assess the antioxidant capacity of extracts (Oloyede *et al*., 2013; Salau *et al*., 2015). Onawumi *et al*. (2012) also reported the presence of saponins and absence of steroids in *S. ocymoides* leaves. The presence of flavonoids in *A. muricata* fruits and its ability to scavenge DPPH radical agrees with reports by Boakye *et al*. (2015). Vijayameena *et al*. (2013) also reported the presence of alkaloids, flavonoids and tannins in the methanol leaf extract of *A. muricata*.The ability of the leaf extracts and fruit juice to reduce these oxidized molecules is an indication of *in vitro* antioxidant capacity, which has been linked to possible *in vivo* antioxidant capacity and ability to prevent oxidative damage to cellular macromolecules such as lipids, proteins and DNA (Oloyede *et al*., 2013; Salau *et al*., 2015).

It should be noted that the methanol extract of *S. ocymoides* leaves appears to possess more phenolics and antioxidant property due to its ability to scavenge DPPH and ferric ions more than the ethyl acetate and n-hexane extracts. The fruit juice of *A. muricata* also had more phenolics and scavenged more of the radicals than its leaf extracts. This is may be due to the fact that the amounts of phenolics and DPPH radical scavenging activity have been found to correlate more with total antioxidant capacity than flavonoids and ascorbic acid content (Olajire and Azeez, 2011) since the phenolic hydrogen is important in the DPPH reaction mechanism.

In conclusion, the results of this study have provided evidence of *in vitro* antioxidant capacity of the hexane, ethyl acetate and methanol leaf extracts of *S. ocymoides* and *A. muricata* as well as *A. muricata* fruit juice. These activities and the various phytochemicals including vitamins present in these extracts might have been responsible for the acclaimed traditional and reported uses of these plants. The plants should therefore be investigated for *in vivo* antioxidant activities.

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