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Characteristics of Fractionated Components of *Aframomum danielli* (Bastard cardamom) Seeds as Natural Food Additives in Soya Oil

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ABSTRACT

Aframomum danielli extracts were obtained using diethyl ether and methanol. The extracts were fractionated using vacuum liquid chromatography. The diethyl ether extraction produced three fractions while methanol produced four fractions. Antioxidant activities of Aframomum danielli fractions were determined by total phenolic content, 1,1-diphenyl,-1,2-picrylhydrazy (DPPH) free radical scavenging power, the rate of inhibition of peroxidation in soya oil and percentage reduction of Free Fatty Acid (FFA). The methanol fraction 3 (MF3) and diethyl ether fractions 2 (DF2) had phenolic contents of 3.64g/100gGAE and 2.5 g/100gGAE respectively. MF3 and DF2 showed antioxidant activities of 84.28% and 55.82% respectively. The DPPH-scavenging power of methanol and diethyl ether fractions ranged from 80.8 to 84.28% and 42.35 to 55.8% respectively. The MF3 showed highest antioxidant activity of 60.24% compared to 55.84 % for DF2 on soya oil after 20 days of storage. The peroxide values and the free fatty acid levels of the soya oil were reduced by all the fractions with increased concentration of Aframomum danielli in soya oil within 20 days of storage. This shows that A.danielli fractions is able to act as a natural antioxidant in the prevention of oxidation of the soya oil.

Key words: *Aframomum danielli,* extracts, components, natural food additives, antioxidant activity

INTRODUCTION

Some of the synthetic chemical additives being currently employed in the food industry for the preservation of foods have been found to be toxic to man (Fasseas *et al.,* 2007, Naveena *et al*., 2008) but natural additives which can be obtained from spices and herbs could serve as non-toxic replacements. Natural preservatives from spices and herbs have been known to have potential for wide applications in extending the storage life of foods (Park, 2011, Shan *et al*., 2005, Wogdylo *et al.,* 2007). Such potential includes antioxidant activity that have resulted from their essential oils, caffeic acid derivatives, flavonoids and terpenoids (Schwarz *et al*., 2001; Tanabe *et al*., 2002, Shan *et al.,* 2005, Flamini *et al.,* 2007).

Moreover, several works have reported the antioxidative from natural sources, such as oilseed, grains, vegetables, fruits, leaves, waxes, bark, roots, hulls and seaweeds (Tuberoso *et al*., 2009; Verma *et al*., 2009; Venkatachalam *et al*., 2014).

Aframomum species as well as other Zingeberaceae are known for the production of Labdane diterpenoids, flavonoids, sesquiterpenoids and arylalkaloids (Tane *et al*., 2002). Erukainure *et al*. (2011) identified flavonoids, tannins, cardiac glycosides, steroids, terpenoids, saponins, alkaloids and phenols in *A. sceptrum*. Besides *A. danielli* was reported to have useful essential oil components (Adegoke *et al.,* 1998).

The antioxidant properties of *A. danielli* and *A. melegueta* have been reported (Fasoyiro *et al.,* 2001, Adegoke *et al.*, 2003). A quinone derivatives from the extract of the seeds of *A. danielli* was isolated by Odukoya *et al.* (1999), which had inhibitory effect on Soya 5-Lipooxigenate. Till date, there exists little information on the fractions derivated from the essential oils obtained from *A. danielli* despite the huge reports on its wide utilization.

Thus, this study was to evaluate the antioxidant capabilities of the essential oils of fractionated components of *A. danielli* extracts as natural food preservative in soya oil.

MATERIALS AND METHODS

Extraction of the Components of *A. danielli*

The seeds of *A. danielli* obtained from Ogbagi Akoko area, Nigeria were removed from the pods, washed in water and air- dried at 27 ± 2°C for three days. The seeds were milled to a coarse particle using a Kenwood blender. One thousand grammes (1000g) of ground seeds was defatted with 2L of petroleum ether (40-60°C) using the method described by Adegoke and Gopala (1998). The residue was then further extracted using diethyl ether and methanol in a Soxhlet apparatus for six hours with special precaution during extraction to reduce solvent vaporization. The extract was concentrated by distilling off the solvent in a rotary evaporator (Laborota 4000, Heidolph, Germany). The extract was further dried in a vacuum oven at 60°C for 24h.

Fractionation of Crude *A. danielli* Extracts using Vacuum Liquid Chromatography

A. danielli crude extracts obtained from diethyl ether and methanol were fractionated using vacuum liquid chromatographic principle as described by Odukoya *et al.* (1999). The vacuum was packed with silica gel as stationary phase and a mobile phase solvents of hexane:ethyl acetate and ethyl acetate:methanol mixtures used in eluting the column in the increasing order of their polarity. The samples were pre-adsorbed using10g of sample dissolved in 50 mL of solvent used in extraction and 50 g of silica gel (Kielselgel 60G Merck) was added. The mixture was dried in a vacuum oven (Gallenkamp) at 60°C to give a free flowing pre-adsorbed sample. Whatman filter paper (125mm) was placed in the chromatographic column and 10 g of the adsorbent silica gel (Kielselgel 60G) was dry packed under vacuum. Another filter paper was placed on the adsorbent and the pre-adsorbent sample was then packed under vacuum in the chromatographic column. The samples were then eluted with different solvents of increasing polarity under vacuum and sucked dry to collect 50 mL each fraction.

Identification of *A. danielli* fractions using Thin Layer Chromatography

The *A. danielli* fractions obtained from crude diethyl ether and methanol were identified by thin layer chromatography (TLC) according to the method described by Haborne (1998) and Hostettman *et al.* (1985). Each fraction was spotted on silica gel $60F_{254}$ TLC aluminium sheets. A mixture of solvent systems of ethyl acetate: hexane (7:3), ethyl acetate: methanol (8:2), ethyl acetate: methanol (7:3) were used in developing the spots depending on the polarity of the fractions. Spots were observed under UV lamp (254nm) and retention factors (R_f) was calculated as the distance travelled by the solute divided by the distance travelled by the solvent for each spots as described by Harborne (1998). Fractions with similar R_f values were bulked together and the samples were concentrated by evaporating on a waterbath as described by Houghton and Raman (1998) and Odukoya *et al.* (1999).

Deterrmination of Peroxides, Acid values and Total phenolic content

Freshly refined soya oil was collected from Sudit Oils and Chemicals, Orita Challenge, Ibadan. The peroxide value in soya oil was determined volumetrically using the methods of Pearsons' (Egan *et al.,* 1991). The decrease in the rate of peroxides was taken as a measurement of the antioxidant activity of each extract. Standard tocopherol was used as positive control. The peroxide value (PV) was determined at five day interval in triplicates and the percentage antioxidant activity (%AOA) was calculated as:

$$\%\text{AOA} = \frac{\text{PV (control)} - \text{PV (test sample)} \times 100}{\text{PV (control)}}$$

The acid value of the stored soya oil was determined using the methods of Pearsons' (Egan *et al*., 1991). The acid value is then expressed as the percentage free fatty acid (%FFA) which is usually calculated as oleic acid. The total phenolic content in *A. danielli* fractions was determined using Folin-Ciocalteu reagent as quoted in Asami *et al.* (2003).Each of the samples (0.05g) was extracted with a 5mL mixture of acetone, water and acetic acid (70: 29.5: 0.5 v/v) and allowed to stand for one hour. The mixture was centrifuged at 1640 rpm for 15 min and the supernatant decanted into polypropylene tubes; the filterate was concentrated in a rotary evaporator (Rotavapor R110, BUCHI, Switzerland) under partial vacuum at 40°C to 2.5 mL. The clear extracts were analysed for phenolic content in triplicates and the absorbance measured using a UV Spectrophotometer (Biotech Novaspec II, Pharmacia, Sweden) at 750 nm. The results were expressed as gramme gallic acid equivalents g(GAE)/100g. Gallic acid equivalents were determined from a standard concentration curve of 5.0 - 30.0 µg/mL Gallic acid.

Determination of Antioxidant Activity

Antioxidant activity in soya oil was determined using the methods of Lee *et. al*. (2003). The extracts were dissolved in 1 ml of 80% ethanol and added directly into the oil at concentrations of 250 and 500 µg/mL. the accelerated Schaal oven storage test was carried out and oxidative stability of treated samples was monitored in an oven at 62° C for 20 days.

DPPH Radical Scavenging of the fractions

Antioxidant activity in *A. danielli* fractions was determined using 2,2,- diphenyl-2-picryl-hydrazyl (DPPH) radical method as described by Aderogba *et al.*(2004) with slight modification. An aliquot of the samples (0.05g dissolved in 100 mL methanol) was prepared into concentrations of 12.5 – 500 µg/mL. About 2.0 mL of each sample was mixed with 1.0 mL solution of DPPH (0.25 mM) in methanol. The decrease in absorbance was measured in triplicate using UV Spectrophotometer (Biotech Novaspec II, Pharmacia, Sweden) at 514 nm. Pure methanol was used to calibrate the spectrophotometer.

Antioxidant activity was calculated as percentage inhibition of the DPPH radical using the equation:

$$AA\% = \frac{\{(\text{Abs sample} - \text{Abs blank}) \times 100\}}{(\text{Abs control})}$$

AA% = Antioxidant activity percent

Data Analysis

All data were obtained in triplicate and means deviations were calculated. The data were subjected to analysis of variance (ANOVA) using the SPSS ver. 15.0 (SPSS, 2006), and the means separated using Duncan multiple range test at 5% probability level.

RESULTS AND DISCUSSION

Table 1 shows the results of the Vacuum Liquid Chromatography (VLC) analysis of diethyl ether extracts. From the VLC, eleven eluents were collected and bulked into three major fractions; diethyl ether fraction 1 (DF1), diethyl ether fraction 2 (DF2) and diethyl ether fraction 3 (DF3). Fractions DF1 was a yellow oil with R_f of 0.70 – 0.71, fraction DF2 was brown solids with R_f of 0.67 – 0.68 while fraction DF3 was a dark brown solids with R_f of 0.64. The result of the VLC for methanol extract, sixteen eluents observed were bulked into four major fractions; methanol fraction 1 (MF1), methanol fraction 2 (MF2), methanol fraction 3 (MF3) and methanol fraction 4 (MF4) using different solvent system are also presented in table 1. Their R_f values ranged from 0.80 - 0.85 (MF1), 0.78 - 0.79 (MF2), 0.75 - 0.76 (MF3) and 0.62 – 0.68 (MF4). The developed colours observed by the subjective assessment shows the fractions MF1, MF2, MF3 and MF4 were a brownish yellow solid, brown solid, dark brown solids and black solid respectively. The result further revealed the dependency of the polarity of the fractions on the nature of the eluting solvents.

Peroxides and Acid values of *A. danielli* in Soya oil

The change in peroxide values and percentage antioxidant effectiveness of *A.danielli* fractions measured by the inhibition of peroxidation in the soya bean oil when

Table 1: Thin layer chromatography of diethyl ether extract and methanol extract of *A.danielli*

Eluting solvent	Nature of oil	Solvent ratio (v/v)	Number of spots	Retention factors
Hexane:EtoAc of up to 70:30 gave DF1	Yellow oil	EtoAc/hexane	1	0.71
		70 : 30	1	0.7
Haxane/EtoAc of up to 30:70 gave DF2	Brown solids	EtoAc/hexane	1	0.67
		70:30	1	0.68
Hexane/EtoAc of up to 10:90 gave DF3	Dark brown solids	EtoAc/hexane 70:30	1	0.64
Toluene/EtoAc of up to 70:30 gave MF1	Brownish yellow solids	EtoAc/MeOH	2	0.80, 0.85
		70:30	1	0.85
Toluene/EtoAc of up to 40:60 gave MF2	Brown solids	EtoAc/MeOH	1	0.79
		40:60	1	0.78
Toluene/EtoAc of up to 10:90 gave MF3	Dark brown solids	EtoAc/MeOH	1	0.76
		80:20	1	0.75
EtoAc/MeOH of up to 50:50 gave MF4	Black solids	EtoAC/MeOH	1	0.68
		80:20	2	0.62, 0.68

Legend: DF1 – Diethyl ether fraction 1; DF2 - Diethyl ether fraction 2; DF3 – Diethyl ether fraction 3; MF1- Methanol fraction 1; MF2- Methanol fraction 2; MF3- Methanol fraction 3; MF4- Methanol fraction 4

Table 2: Change in peroxide value in soya oil during storage using *A. danielli* fractions at 500 µg/mL

Sample	DAY 0	DAY 5	DAY 10	DAY 15	DAY 20
DE	1.33	18.95±0.18^{cd}	29.62±0.33^{cd}	47.64±0.43^{f}	52.67±0.41^{d}
DF1	1.33	19.15±0.13^{cd}	29.80±0.53^{d}	49.25±0.25^{g}	55.33±0.76^{f}
DF2	1.33	18.20±0.10^{ab}	27.80±0.27^{b}	43.55±0.83^{c}	50.34±0.71^{c}
DF3	1.33	20.54±0.50^{e}	32.21±0.84^{f}	50.99±0.34^{h}	57.67±0.38^{g}
ME	1.33	19.11±0.16^{cd}	29.09±0.25^{cd}	45.83±0.1^{de}	54.60±0.40^{ef}
MF1	1.33	18.74±0.26^{bc}	29.12±0.59^{cd}	46.60±0.3^{ef}	54.67±0.50^{ef}
MF2	1.33	17.98±0.17^{ab}	26.96±0.20^{b}	42.50±0.70^{b}	47.83±0.35^{b}
MF3	1.33	17.29±0.46^{a}	23.64±0.15^{a}	41.33±0.16^{a}	45.33±0.76^{a}
MF4	1.33	18.44±0.12^{bc}	28.76±0.12^{c}	45.43±0.61^{d}	53.50±0.89^{de}
Tocopherol	1.33	19.85±0.13^{de}	31.35±0.18^{e}	49.48±0.46^{g}	58.33±0.59^{h}
Oil control	1.33	34.13±2.01^{f}	55.67±1.53^{g}	93.33±1.15^{i}	114.0±1.60^{i}

Mean± Standard deviation. Values in the same column with different superscript are significantly different ($p \leq 0.05$)

Table 3: Change in peroxide value in soya oil during storage using *A. danielli* fraction at 250 µg/mL

Sample	DAY 0	DAY 5	DAY 10	DAY 15	DAY 20
DE	1.33	20.32±0.35^{bc}	31.45±0.82^{cd}	50.60±1.05^{de}	56.82±0.23^{f}
DF1	1.33	20.75±0.28$_{cd}$	32.14±0.41^{de}	52.18±0.75^{f}	56.72±0.23^{ef}
DF2	1.33	19.56±0.63^{ab}	30.55±0.90^{cd}	48.50±0.34^{bc}	54.3±0.48^{bc}
DF3	1.33	23.14±0.15^{f}	35.11±1.34^{f}	53.20±1.06^{f}	60.48±0.29^{g}
ME	1.33	21.40±0.36^{de}	30.39±0.28^{bc}	48.97±0.51^{cd}	54.75±0.25^{cd}
MF1	1.33	20.36±0.56^{bc}	31.37±0.60^{cd}	49.89±0.20^{de}	55.96±0.53^{de}
MF2	1.33	19.74±0.26^{ab}	29.73±0.30^{ab}	47.80±0.44^{b}	53.47±0.67^{b}
MF3	1.33	18.95±0.84^{a}	28.67±0.57a	44.30±0.46^{a}	50.57±0.25^{a}
MF4	1.33	20.19±0.51^{bc}	31.48±0.46^{cd}	49.60±0.46^{cd}	55.30±0.46^{cd}
Tocopherol	1.33	20.61±0.32^{bc}	32.70±0.76de	52.80±0.27^{f}	60.67±0.45^{g}
Oil control	1.33	34.13±2.01	55.67±1.53	93.33±1.15	114.0±2.0

Mean± Standard deviation. Values in the same column with different superscript are significantly different ($p \leq 0.05$)

fractions were added to the oil at 250 and 500 µg/mL is shown in Tables 2 – 4 while the percentage free fatty acids are shown in Figures 1 and 2. *A.danielli* antioxidant extracts showed reduction in rancidity compared to the untreated soya oil. Oxidative stability in soya oil was increased by the rate of decrease in percentage free fatty acid formation. Meanwhile, the control peroxide values and free fatty acid increased with storage time in the soya oil while the peroxide value and free fatty acid levels decreased with increased *A. danielli* concentrations. A significant difference ($p \leq 0.05$) was observed in the peroxide values of the fractions and the soya oil control. All the fractions of *A.danielli* exhibited antioxidant activities with methanol fraction 3 (MF3) found to possess the highest activity of 60.24% followed by MF2 (58.04%), MF4(53.07%), MF1(52.04%) and DF2 (55.84%) after 20days of storage at 500µg/ml (Table 4). The result is very encouraging compared to the activity of α–tocopherol being used as control in soya oil that showed 48.83% antioxidant effectiveness. Thus, the antioxidant activity of the fractions can be attributed to the phenolic contents in the fractions whereas the prevention oxidation that leads to rancidity in soya oil is ascertained by the use. Phenolics are known to be effective in preventing lipid oxidation, thereby increasing oil stability in foods (Bopitiya and Madhujith, 2013).

The order of action of antioxidant property for methanol fractions was MF3> MF2>MF4>MF1, while that of diethyl ether fractions was DF2> DF1>DF3 (Table 4).

Adegoke *et al.* (2003) reported the antioxidant effectiveness of *A. melegueta* on lard (57%) and groundnut oil (53.43%) at 300ppm compared with 29.6% for *Xylopiaaethiapica*. Adegoke *et al.* (2000) also reported a 70.7% at 200ppm as crude antioxidant activity from *A.danielli* compared with α–tocopherol of 46.1% activity. Fasoyiro *et al.* (2001) reported *A.danielli* with higher antioxidant activity than Stabex and tocopherol. Amarowiez *et al.* (2000) also observed high antioxidant activity in rape seed and Canola fractions. This shows that extracts from plant materials possess antioxidant property which could found functional application in food systems.

Table 4: Antioxidant activity (AOA%) of *Aframomum danielli* fractions in soya oil during storage

Sample	Concentration	
	250 µg/mL	500 µg/mL
DE	50.16±0.20^{g}	53.80±0.35^{d}
DF1	50.25±0.20^{g}	51.46±0.67^{ef}
DF2	52.33±0.42^{c}	55.84±0.61^{c}
DF3	46.95±0.25^{h}	49.41±0.33^{g}
ME	51.97±0.22^{cd}	52.11±0.36^{e}
MF1	50.91±0.46^{ef}	52.04±0.44^{e}
MF2	53.10±0.58^{b}	58.04±0.94^{b}
MF3	55.64±0.22^{a}	60.24±0.69^{a}
MF_4	51.49±0.40^{de}	53.07±0.78^{d}
Tocopherol	46.78±0.40^{h}	48.83±0.52^{h}

Mean± Standard deviation. Values in the same column with different superscript are significantly different ($p \leq 0.05$)

Total Phenolic content of *A. danielli* fractions

The distribution of phenolic compounds in *A. danielli* fractions using the Folin Ciocateu testis shown in Table 5. The result shows that the highest level of total phenolic content 3.64g/100g GAE was found in the methanol fraction 3 (MF3) followed by MF2, MF1 and MF4 (3.32, 3.14 and 2.92 g/100g GAE) respectively, while the diethyl fraction 2 (DF2) had 2.51g/100g GAE. The result shows significant difference ($p \leq 0.05$) in the distribution of phenolics from *A.danielli* fractions using different extracting solvents whereas methanol seemed to be the best extracting solvent for phenolic components in *A.danielli.*

The total phenolic content obtained from *A. danielli* shows that an inclusion of *A. danielli* fractions in food would increase the antioxidants content and would have potential as a natural antioxidant which could thus inhibit unwanted oxidative processes. The values for total phenolics of all the fractions compared favourably with ascorbic acid values of 2.94g/100g. GAE. The present result is lower than 5.45g/100g GAE reported for antioxidant activity of extracts from coriander leaves but higher than 1.89g/100g GAE for its seeds (Wungesteen *et al*., 2004). Phenolic compounds have been reported to have high antioxidant activity by scavenging the free radicals in the body (Zheng *et al*., 2009). Antioxidant activities of foodstuff have been reported to significantly increase with the presence of high concentration of total phenol content of such food (Jayaprakasha *et al*., 2008).

Table 5: Total Phenolic content of *Aframomum danielli* fractions

SAMPLE	g (GAE)/100g
DE	2.39 ±1.90^{g}
DF1	2.12 ± 0.25^{h}
DF2	2.51 ±2.10^{f}
DF3	2.08 ± 0.75^{i}
ME	3.22 ± 1.37^{c}
MF1	3.14 ± 0.90^{d}
MF2	3.32 ± 1.51^{b}
MF3	3.64 ± 1.31^{a}
MF_4	2.92 ±0.45^{e}
Ascorbic Acid	2.94 ± 0.85^{e}

Mean± Standard deviation. Values in the same column with different superscript are significantly different ($p \leq 0.05$)

DPPH Radical Scavenging activity of *A. danielli* fractions

Table 6 shows the radical scavenging activity of *A.danielli* fractions. There was significant difference ($p \leq .05$) between the scavenging abilities of the methanolic fractions compared with the diethyl ether fractions. The methanolic extracts and fractions from *A. danielli* showed the highest DPPH radical scavenging abilities of 80.8 - 84.28% at 500 µg/mL. The diethyl ether fractions had DPPH radical scavenging abilities of 42.35 - 59.42% when compared with the ascorbic acid standard 26.56 - 92.84%. at 2 – 10 µg/mL concentration. Similar trends were reported by Mau *et al.* (2004) of the methanolic extracts from '*ling chih* and baby *ling chih'* that showed high DPPH radical scavenging abilities of 88.4% and 93.8% at 5 mg/ml, respectively. Besides the methanolic extracts from mycelia and filtrate scavenged DPPH radicals by 85.7 % and 79.3% at 10 mg/mL, respectively. It was reported that at 0.1mg/ml BHA and α-tocopherol showed excellent scavenging abilities of 99.9% and 95.1% (Mau *et al.,* 2004). Huang *et al.*(2002) found that the

Table 6: Percentage antiradical activity (AA%) of *A. danielli* fractions using DPPH scavenging

Sample	Concentration (mg/mL) of *A. danielli*					
	500	**250**	**125**	**50**	**25**	**12.5**
DE	59.42±0.82^{c}	31.29±0.81^{e}	23.13±0.90^{e}	18.23±0.88^{cd}	17.72±0.56^{e}	15.72±0.27^{d}
DF1	50.33±0.93^{e}	25.45±0.61^{g}	20.31±0.73^{f}	18.50±0.44^{cd}	16.73±0.34^{f}	14.88±0.42^{e}
DF2	55.82±0.57^{d}	27.96±0.95^{f}	23.27±0.73^{e}	18.64±0.63^{c}	16.27±0.74^{f}	15.04±0.12^{e}
DF3	42.35±0.82^{f}	27.65±0.50^{f}	23.66±0.68^{e}	17.96±0.22^{cd}	16.64±0.34^{f}	15.41±0.42^{de}
ME	81.42±0.61^{b}	74.69±0.53^{c}	70.56±0.60^{a}	55.65±0.59^{a}	34.24±0.80^{c}	22.85±0.54^{b}
MF1	81.28±1.52^{b}	75.39±1.17^{bc}	68.72±0.46^{bc}	54.82±0.55^{a}	37.44±0.70^{b}	21.73±0.37^{c}
MF2	83.63±0.87^{a}	76.37±0.77^{ab}	69.28±0.39^{b}	55.43±0.66^{a}	38.48±0.63^{a}	23.92±0.55^{a}
MF3	84.28±1.12^{a}	76.68±0.48^{a}	68.14±1.08^{c}	55.68±0.42^{a}	34.46±0.86^{c}	23.46±0.51^{a}
MF4	80.8±0.35^{b}	72.16±0.31^{d}	64.43±0.61^{d}	49.59±0.45^{b}	30.37±0.80^{d}	22.34±0.35^{b}

	Concentration (mg/mL) of *A. danielli*				
	10	**8**	**6**	**4**	**2**
AA	92.84±0.84^{a}	75.83±0.62^{b}	59.60±0.65^{c}	43.10±0.55^{d}	26.56±0.34^{e}

Mean with different superscripts are significantly different ($p<0.05$).

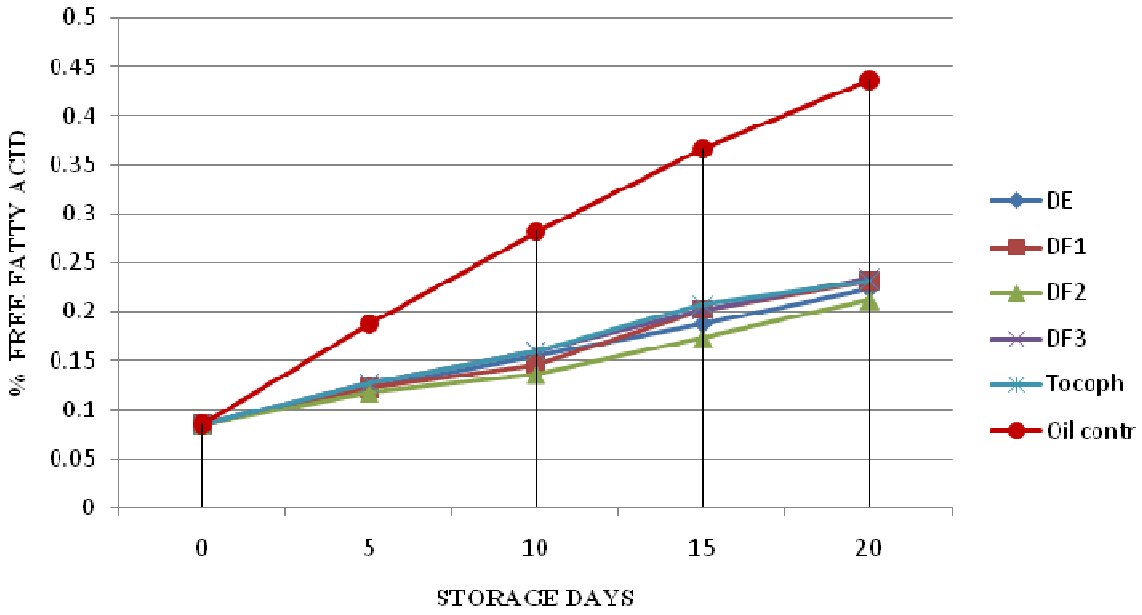

Figure 1: Percentage Free Fatty Acid using 500µg/mL Diethyl Ether Fractions of *A. danielli* in Soyabean Oil

scavenging ability of the methanolic extract from *A. cylindracea* fruit bodies at 1mg/ml was 89.0%.

The fraction DF3 has the same scavenging ability value (42.3 % at 500 µg/mL) with that obtained for *ganoderma tsugae* extracts (Mau *et al* 2005). The total phenolic contents of *A. danielli* fractions (Table 5) may be responsible for the observed high scavenging ability of the fractions. Zhou and Yu. (2006) reported the correlation between the total phenolic content of the tested vegetable extracts and its DPPH radical scavenging activity, suggesting that total phenolics could play a major role in the antioxidant activity of plant materials.

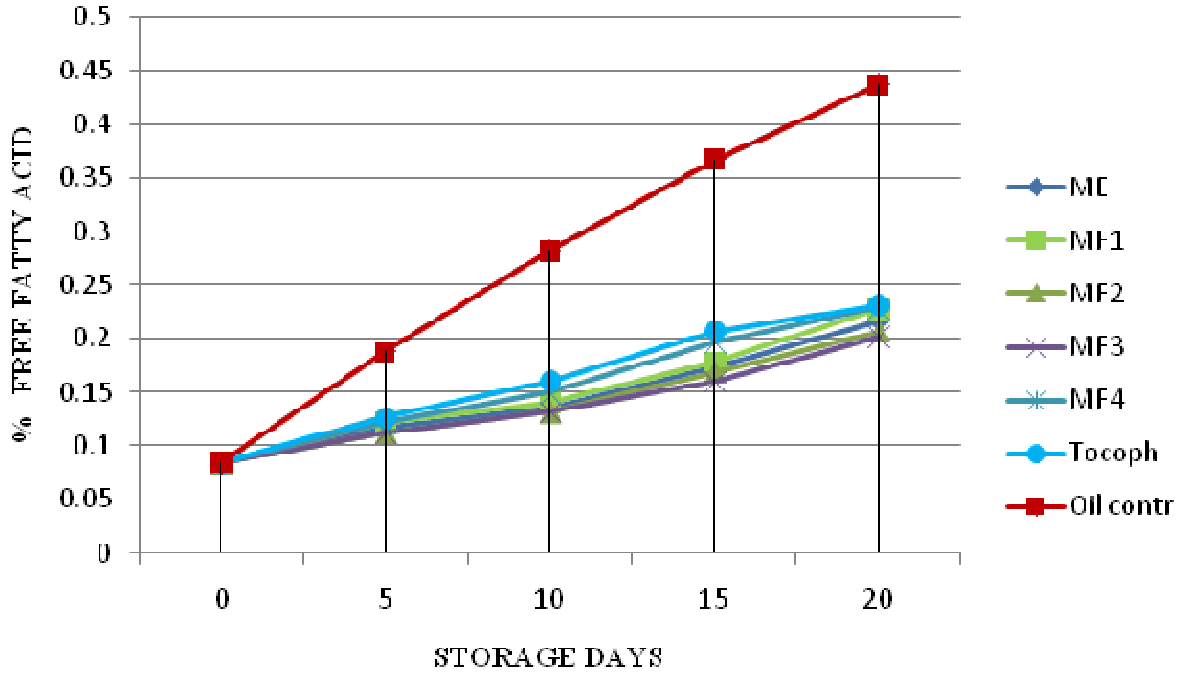

Figure 2: Percentage Free Fatty Acid using 500µg/mL Methanol Fractions of *A. danielli* in Soyabean Oil

CONCLUSION

This study has shown the characteristics of *A.danielli* fractions obtained from diethyl ether and methanol extracting solvents. It has shown that essential oils from *A. danielli* fractions possess antioxidative property in soya oil. This is an indication that *A.danielli* fractions promises to be an excellent natural source of food preservatives that could be utilized in the food industry.

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