



Research article

Effect of *Syzigium aromaticum* and *Allium sativum* spice extract powders on the lipid quality of groundnuts (*Arachis hypogaea*) pudding during steam cooking

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ABSTRACT

Groundnut seeds (*Arachis hypogaea*) contain higher concentrations of unsaturated lipids which are prone to oxidation in formulated foods. This study determined the antioxidant activities of water extract powders from two spices (*Syzigium aromaticum* and *Allium sativum*) and their ability to preserve the quality of lipids in groundnuts pudding during steam cooking with 0, 0.5, 1, 2 and 4% of spice extract powders. Total phenolic (TPC) and flavonoid (FC) contents of extracts from *S. aromaticum* were 140.23 mg GAE/100g extract and FC of 110.34 mg CAE/g extract compared to values of *Allium sativum* extracts (54.28 mg GAE/100g extract and 34.80 mg CAE/g extract). The showed DPPH free radical scavenging activities of the extract from *S. aromaticum* depending on the concentration ranged from 82.15% to 97.66% and this was higher than the activities of *A. sativum* but comparable to the values of butylhydroxytoluene used as control. The chemical analysis of oil extracted revealed that the addition of the spice extract powders limited the appearance of oxidation products characterized by a reduction of up to 9-fold of peroxide value, 5-fold for anisidine and 2-fold for thiobarbituric acid reactive species. In many cases, the addition of *S. aromaticum* spice extract powder to the pudding better prevented lipid oxidation likely because of its superior ability to scavenge peroxy radicals (ROO[•], HO[•], DPPH[•]). In a nutshell, the addition *S. aromaticum* and *A. sativum* spice extract powders on grilled groundnuts paste for groundnuts pudding preparation in household can help preserve its lipid quality.

1. Introduction

Oilseed are important crops for population worldwide, as they can provide both oil and quality proteins. Their lipids provide nutritional, sensory and technological functions to foods (Rios et al., 2014). The major oilseeds been used by housewives in Cameroon for soup preparation is groundnuts because it is very cheaper and it can be affordable by all social classes level (Kengap et al., 2019). Groundnut seeds are rich sources of macronutrients such as lipids (46.22%), proteins (25.26%) depending on the variety and carbohydrates (21.26%) that can be soluble or non soluble. They also contained micronutrients such as vitamins and minerals (Ingale and Shrivastava, 2011). The Groundnut oil is rich in monounsaturated (palmitoleic and oleic acids) and

polyunsaturated fatty acids such as linoleic and α -linolenic acids (Aluyor et al., 2009). These two polyunsaturated fatty acids are precursors of hormone biosynthesis such as prostaglandins. However, after harvest, groundnut seeds are subjected to many cooking technique which are likely to reduce their nutrients content. In fact, groundnut seeds contain about 79.59% of unsaturated fatty acids which are prone to degradation (oxidation and decomposition) during heat treatment (Aluyor et al., 2009). The extent of oxidation is affected by temperature, time, and the presence or absence of water.

Groundnuts pudding is a meal that is been prepared with roasted groundnuts paste in which spices are added, tied in banana leaves and that undergo steam cooking before consumption. This meal is rich in proteins (18.30%); lipids (35.00%); 11.50% of available carbohydrate,

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minerals and it is widely consumed in the centre region in Cameroon (Kouebou et al., 2013; Sharma et al., 2007). The type of spices added, the possible combinations and quantities usually vary according to availability, preferences, traditional indications and the financial means of households. Previous works such as those of Womeni et al. (2013) have shown that *Syzigium aromaticum* and *Allium sativum* have antioxidant properties, as determined from extracts with solvents such as ethanol and methanol. Although, these solvents have higher capacity to extract polyphenols, for obvious reasons, they are not part of groundnuts pudding preparation where spices and groundnuts paste are mixed with water. A survey carried out on groundnut pudding sellers in few cities in Cameroon helped us to identify *Syzigium aromaticum* commonly called clove and *Allium sativum* commonly called garlic as the most used spices for groundnut pudding preparation. Previous works such as those of Santhosha et al. (2013) have showed that *Allium sativum* contains strong antioxidant compounds such as S-allylcysteine and S-allyl mercaptocysteine. *Allium sativum* is able to inhibit lipid peroxidation and LDL oxidation thus contributing to decrease the risk of cardiovascular disease (Lau, 2006). Many authors have also shown that *Syzigium aromaticum* possess antioxidant activity and this activity is attributed to its high level of phenolic compounds (Dudonne et al., 2009; Womeni et al., 2013; Roman et al., 2015). The interest in the present study was to determine how the difference in either the concentration of a water extract from the spices in dried and powdered form, their phenolic contents, or their antioxidant activity could affect the oxidative status of lipid as characterized by primary and secondary oxidation products.

2. Material and methods

2.1. Collection of spices

Two spices (2) *Syzigium aromaticum* (Z.A) and *Allium sativum* (A.S) and dry groundnuts seeds were bought in Dschang market, Menoua's Head Division and identified at the National Herbarium Yaounde in Cameroon.

Peeled *A. sativum* and whole *S. aromaticum* seeds were dried in an oven at 55 °C for 48 h. The dried spice was ground to less than 1 mm using a local fabrication mill that was bought from a trader in Dschang market in order to obtain a powder.

2.2. Extracts preparation

Twenty-five grams of each powder from the two spices were extracted into 500 ml of water for 48 h with regular manual shaking after every 3 h at room temperature and we were sleeping in the laboratory for the experiment to be well carried out (Womeni et al., 2013). The moisture content of *A. sativum* powder used for extract preparation was 8.59% and that of *S. aromaticum* was 5.21%. The extracts were then filtered using Watman No. 1 filter paper and the filtrates placed in an oven at 45 °C for 48 h for partial removal of the remaining solvent. The extracts obtained were stored at 4 °C for further analysis. After drying at 40 °C the residue was a tight cake, not crystalline and soluble in reagent after being diluted with distilled water according to the test that was to be carried out.

2.3. Determination of total phenolics content (TPC)

The total phenolics content (TPC) was determined using the Folin Ciocalteu colorimetric method as described by Gao et al. (2000). For this experiment, 20 µl of Spice powders extracts with concentration 2 mg/ml were mixed in a test tube with 0.2 ml of Folin-Ciocalteu reagent plus 2 ml of distilled water and incubated at room temperature for 3 min. One ml of 20% sodium carbonate was added to the mixture, which was re-incubated for 2 h at room temperature. The absorbance of the resulting blue color was measured on a BIOMATE brand spectrophotometer (Thermo Scientific™ 840208400/EMD) using a quartz cuvette at 765 nm. The standard curve was prepared from Gallic acid solution at

concentration of 0.2 g/l and used to express the results as mg gallic acid equivalents (GAE) per gram of spice extract powder.

2.4. Determination of flavonoids content (FC)

The flavonoids content (FC) was determined using the methods described by Marinova et al. (2005). Spice powders extracts (0.1 ml) at concentrations of 2 mg/ml were mixed with 1.4 ml distilled water and 0.03 ml of sodium nitrate solution (NaNO₃) (5%) was added. After 5 min 0.2 ml of 10% aluminum trichloride (AlCl₃) was added. 0.2 ml of 10% sodium hydroxide (NaOH) and 0.24 ml of distilled water was also added after 5 min. The solution was then shaken and the absorbance measured on a BIOMATE brand spectrophotometer using a quartz cuvette at 510 nm. The standard curve prepared from catechin at concentration of 0.1 mg/ml was used and the flavonoid content (FC) expressed as mg catechin equivalents (CAE) per gram of extract.

2.5. Evaluation of the antioxidant activity by the DPPH free radical scavenging assay

The DPPH radical scavenging ability of the spice extracts was determined according to the method of Mensor et al. (2001). For this, 1 ml of 0.3 mM methanolic solution of DPPH was added to 2.5 ml of the samples at different concentrations (250, 500, 1000 and 2000 µg/mL). Mixtures were kept at room temperature in the dark for 30 min after which, the optical density was measured at 517 nm on a BIOMATE brand spectrophotometer (Thermo Scientific™ 840208400/EMD). The absorbance of all samples was measured against a blank. A synthetic antioxidant, butylhydroxytoluene (BHT) prepared at concentrations of 2 mg/ml was used as positive control. The radical scavenging activity (AA) was determined using the following formula:

$$A.A(\%) = \frac{(\text{OD of DPPH} - \text{OD of sample})}{\text{OD of DPPH}} \times 100 \quad (1)$$

2.6. Evaluation of ferric reducing antioxidant power (FRAP)

The reducing power of the extracts was determined by their ability to reduce Iron (III) to Iron (II) as described by Oyaizu (1986).

In test tubes containing 0.5 ml of spice extract powder at 250, 500, 1000 and 2000 µg/mL prepared in distilled water, 1 ml of a phosphate buffered saline solution potassium (0.2 M, pH 6.6) and 1 ml of 1% aqueous potassium hexacyanoferrate [K₃Fe (CN)₆] solution were added. The whole solution was incubated for 30 min at 50 °C in a water bath and 1 ml of 10% trichloroacetic acid was added. The mixture was centrifuged at 3000 g for 10 min and 1.5 ml of supernatant was removed and mixed with 1.5 ml of distilled water; this was followed by the addition of 0.1 ml of 0.1 % FeCl₃ aqueous solution. Butylhydroxytoluene (BHT) at concentrations of 2 mg/ml prepared under the same conditions was used to compare the reducing power of spice extracts. The blank was made of all reagents except the extract and water was not added in place of the spice extract powder. The optical density of the samples and that of the control were measured at 700 nm against this blank on a BIOMATE brand spectrophotometer (Thermo Scientific™ 840208400/EMD). An increase in the absorbance of the reaction mixture indicated an increase in reducing power of the spice extracts to reduce Iron (III) to Iron (II).

2.7. Evaluation of hydroxyl radical inhibition power (ROH)

The capacity of the extracts to inhibit the hydroxyl radical production was done by the method described by Nagulendran et al. (2007).

In test tubes 60 µl of FeCl₂ (1 mM), 90 µl of 1,10-phenanthroline (1 mM), 2.4 ml of 0.2M phosphate buffer (pH 7.8), 150 µl of 0.17 M hydrogen peroxide and 1.5 ml of spice extract powder at 250, 500, 1000 and 2000 µg/mL were mixed. The reaction was initiated by the addition of hydrogen peroxide and incubated at room temperature for 5 min. After

incubation, the absorbance of the mixture was read at 560 nm using a BIOMATE brand spectrophotometer (Thermo Scientific™ 840208400/EMD) and a blank prepared under the same conditions. Butylated hydroxytoluene (BHT) at concentrations of 2 mg/ml was used as a standard and the experiments were performed in triplicate. An increase in the absorbance of the reaction mixture indicated an increase in the ability to reduce the hydroxyl radical.

$$\text{ROH} (\%) = \frac{\sum \text{OD}}{n} \times 100 \quad (2)$$

2.8. Oxygen radical absorbance capacity (ORAC)

The peroxy radical (ROO•) scavenging activity of extracts was determined according to a previously reported procedure (Ratnasari et al., 2017). Samples and standard were made in potassium phosphate buffer (75 mM, pH 7.4). Spice extract powders were diluted to concentrations of 0.2 mg/mL while the concentrations of the standard (Trolox) were 5–100 μM. In a 96-well black microplate, 120 μL of fluorescein (0.08 μM) were added followed by 20 μL of diluted extract or standard. The plate was then sealed and incubated for 20 min at 37 °C in the FLx800 BioTek fluorimeter (BioTek Fisher Scientific, Nepean, ON, Canada). After incubation, 60 μL of AAPH (150 mM) were added to all the 96-well of the black microplate and data were recorded at 1 min interval for a total of 50 min. Net area under the curves were used to calculate ORAC values as μM Trolox equivalents (TE)/g extract.

2.9. Preparation of groundnuts pudding and extraction of oil

Dried groundnuts were ground with an electric blender (Royalty line; Model No: SME-600.6; Order No: 16-RL-942) to a paste and roasted for 10 min at 190 ± 10 °C inside a pot placed on an electric heater. 100 g of paste was mixed with 30 ml of warm water (50 ± 2 °C). The spice extracts from *S. aromaticum* or *A. sativum* were added at 0, 0.5, 1, 2, and 4% respectively. The pastes with and without supplementation with the spice extract were tied in banana leaves and cooked separately for 90 min in a pot placed on an electric heater at 93.5 ± 2 °C. The pot called “Cocotte” in French used for the pudding preparation was a 5L capacity; diameter 27 cm bought in Dschang market from a trader. The length of each pudding tied in banana leaves was about 13 cm. The bottom of the pot was covered with plantain leaves in order to prevent water entry and burning of pudding during cooking. After cooking the groundnuts pudding was allowed to cool at room temperature for 30 min.

2.10. Oil extraction

At the end of cooking by steaming, the oils of different groundnuts pudding were extracted by the method described by Bligh and Dyer (1959). 100 g of groundnuts pudding were introduced into a glass blender (Royalty line; Model No: SME-600.6; Order No: 16-RL-942). 200 ml of a chloroform/methanol (2/1 v/v) mixture were subsequently added into it. After grinding at speed 4 for 10–15 min, 100 ml of additional chloroform was added. After homogenizing the mixture for about 1 min, the homogenate was filtered with Whatman paper No.1, and the resulting filtrate was then placed in a separatory funnel to separate the phases. The lower phase consisting of chloroform/oil mixture was collected and then evaporated at 50 °C on a rotary evaporator, in order to separate the oil from the solvent. The oils obtained were put in dark glass bottles and stored for 48 h in a freezer for subsequent analysis.

2.11. Chemical characterization of lipids extracted from groundnuts pudding after steaming cooking

Lipid quality was evaluated by determining the peroxide value (PV) using the IDF 74A:1991 standard spectrophotometer method (International Dairy Federation Standards, 1991), the anisidine value (AnV)

using the official AOCS Cd 18–90 «*p*-anisidine value» (AOCS, 2003), the thiobarbituric acid value (TBARS) using the method described by Draper and Hadley (1990) and the iodine value (IV) using the AOCS Cd 1–25 official method (AOCS, 2003). The FT-IR spectrometer (Model Spectrum 65; PerkinElmer, Waltham, MA, USA) was used to collect FT-IR spectra with a resolution of 4 cm⁻¹ at 64 scans.

2.12. Statistical analysis

Results were subjected to analysis of variance (ANOVA) with Student Newman-Keuls Multiple Comparison Test, to determine the statistical significance of the data expressed as mean ± standard deviations. A probability value of *p* < 0.05 was considered statistically significant using GraphPad Instat version 5.0 software. In addition, Pearson's Correlation between the total phenolic content (TPC), flavonoid content (FC), free radical scavenging (DPPH), ferric reducing antioxidant power (FRAP), hydroxyl radical (ROH), Oxygen radical absorbance capacity (ORAC), peroxide value (PV), *p*-anisidine value (AnV), thiobarbituric acid value (TBARS) and iodine value (IV) was calculated using SPSS version 20.

3. Results and discussion

3.1. Total phenolic content (TPC) and flavonoids content (FC)

The total phenolic and flavonoid contents of *S. aromaticum* (ZA) and *A. sativum* (AS) extract powders is presented in Table 1. It appears that, TPC and FC were greater for extracts of *S. aromaticum* (ZA) than those of *A. sativum* (AS). The results obtained for the TPC in this study with ZA was higher than that of Milda and Ambuscado (2015) (113.19 mg GAE/g extract). The results obtained with ZA and AS were greater than those reported by Womeni et al. (2013) on methanolic extract of these spices with respective values of 12.35 and 1.34 mg GAE/g extract. The TPC of ZA obtained in this study is significantly less than that of Dudonne et al. (2009) with mean of 212.85 mg GAE/g extract. According to Shan et al. (2005), the differences in the level of the total phenolics and flavonoids contents may be due to factors like climate, location, temperature, fertility, diseases, parts of plant tested, time of taking samples, determination methods and the type of solvent use for extractions. The spices used here are from commercial sources and no information on their growing conditions was available.

3.2. Antioxidant activity of extracts

3.2.1. DPPH free radical scavenging assay

Figure 1(a) illustrates the free radical scavenging activity of the spice extract powder. It can be seen that the antioxidant activity depends on the concentration of extracts (*p* < 0.05). At the lowest tested concentration of 25 μg/ml, the extract of ZA scavenged DPPH radical 82.2 ± 0.01% compared to only 16.0 ± 0.07% of the extract of AS. As compared to scavenging power of AS spice extract powder at 25 μg/ml, the scavenging power of AS extract almost triple to 40 ± 0.00% at maximum concentration (200 μg/ml) while there was only a minimal change for ZA extract because its maximum quenching was reached with lower concentration at 50 μg/ml. The higher TPC content of the ZA extract powder, about 2.7-fold that of AS extract, might explain its higher DPPH radical scavenging activity. The DPPH activity of ZA at each concentration closely matched that of the control BHT and this is in line with previous data with methanolic and aqueous extract of the same spices (Womeni et al. (2013); Dudonne et al. (2009)).

3.2.2. Ferric reducing antioxidant power (FRAP) and inhibition of hydroxyl radical production

Figure 1(b) and figure 1(c) showed the FRAP and the inhibition of hydroxyl radical production of ZA and AS extracts at different concentration compared to that of BHT respectively. At first glance ZA extract

Table 1. Total phenolic content (TPC) and flavonoid contents (FC) of *S. aromaticum* and *A. sativum* extract powders.

Samples	TPC (mg GAE/g)	TFC (mg CE/g)
<i>A. sativum</i>	54.28 ± 0.00 ^b	34.80 ± 0.17 ^b
<i>S. aromaticum</i>	140.23 ± 0.00 ^a	110.34 ± 0.21 ^a

Values with different superscripts differ significantly at $p < 0.05$.

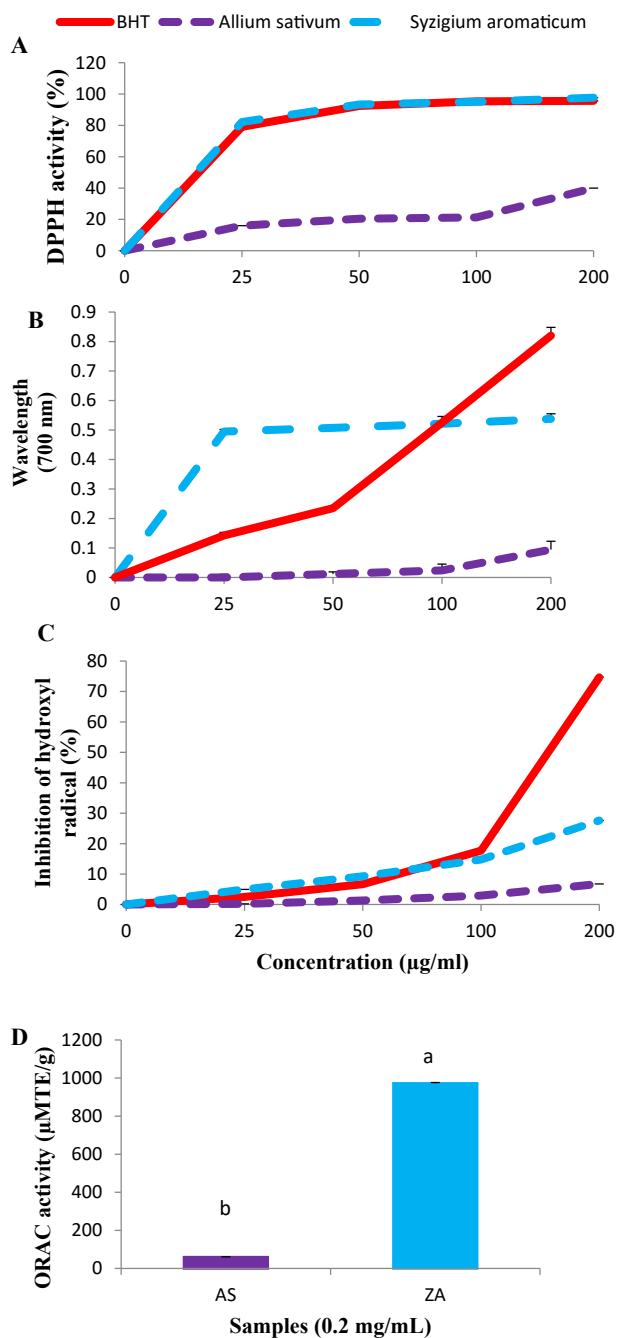


Figure 1. Antioxidant activity of aqueous extracts from *S. aromaticum* and *A. sativum*. (A) DPPH free Radical Scavenging activity. (B) Ferric Reducing Antioxidant Power (FRAP). (C) Inhibition of hydroxyl radicals. (D) Oxygen radical absorbance capacity (ORAC).

showed higher FRAP and inhibition of hydroxyl radical activities ($p < 0.05$) than BHT at 25 and 50 µg/ml respectively. At 100 µg/ml the FRAP and hydroxyl radical inhibition activities are similar ($p > 0.05$) to

that of BHT and ZA extract powder and significantly lower ($p < 0.05$) at 200 µg/ml. However, AS extracts presented a FRAP activity significantly lower ($p < 0.05$) than that of BHT. The highest FRAP activity observed with ZA extract may be due to the presence of hydroxyl group of phenolic compounds such as flavonoids that can serve as electron donors, hereby reducing Iron (III) to Iron (II) (Siddhuraju and Becker, 2007). An increase of the quantity of Iron (II) leads to the increase of the reducing power of the extract. Therefore, antioxidants are considered as reducing and inactivators of oxidants (Siddhuraju and Becker, 2007). These results were in harmony with those of Dudonne et al. (2009) who stated that the aqueous extract of ZA has an iron reduction power higher than the aqueous extract of some plants and spices.

3.2.3. Oxygen radical absorbance capacity (ORAC)

The oxygen radical absorbance capacity (ORAC) of ZA and AS extracts at 0.2 mg/mL is presented in Figure 1(d). ZA (971.95 µMTE/g) presented an ORAC value significantly higher ($p < 0.05$) than that of AS (59.63 µMTE/g). It appears that the ZA extract peroxy radical quenching capacity (ROO[•]) was about 13-fold higher than that of the AS extract. It is known that the ORAC test measures the potential of chemical compounds to donate proton to ROO[•] and that some of the molecules that can easily donate protons are polyphenols (Esfandi et al., 2019). In this regard, the difference in ORAC activities can be explained in part by the TPC and FC on the extracts as TPC and FC values of the ZA extract were about 2.6 and 3.2-fold higher compared to corresponding values for the AS extract. The polyphenolic composition of the extracts as well as their structures likely played a role in their ROO[•] scavenging activities (Esfandi et al., 2019). The ORAC value obtained in this study with ZA is inferior to that of Xinyan et al. (2015) with the aqueous extract of the same spice (8764.78 µMTE/g).

3.3. Effect of added spices on the lipid quality of groundnuts pudding after steam cooking

3.3.1. Peroxide value

The peroxide value (PV) gives information on the primary oxidation state of oil and fats marked by the formation of hydroperoxides. High peroxide values generally reflect oil alteration as a result of oxidation of its unsaturated fatty acids and hydroperoxide formation (Sultana et al., 2008). The effects of ZA and AS extract powders were tested at four concentrations (0.5–4%). It appears that the PV obtained with oils extracted from groundnuts pudding supplemented with 0.5, 1, 2 and 4% of AS and ZA extract powder were significantly lower ($p < 0.05$) than the PV of oil extracted from the control groundnuts pudding prepared without extract powder (Table 2). For groundnuts pudding cooked with AS powder, the lower PV was observed at 0.5 and 1.0% inclusion, while for those cooked with ZA the lower PV was observed with 0.5% inclusion. The reduction of PV of oil from samples treated with AS and ZA spice extract powders may be due to the presence of natural antioxidants in these extracts that may have limited the formation of hydroperoxides since these extracts are rich in phenolic compounds and antioxidant activity (DPPH free radical scavenging and ferric reducing antioxidant power (FRAP)) in varying proportions (Roman et al., 2015). This reduction can also be due to the transformation of hydroperoxides which are unstable substances, to secondary oxidation product responsible for the change in odour and colour (Shermer, 1990). All oil samples extracted from groundnuts pudding supplemented with AS and ZA spice extract powders with the exception of that extracted from groundnuts pudding cooked with 2% ZA extract powder showed PV lower than the recommended Codex Alimentarius value which is 10 meqO₂/kg (Codex, 2015). However, it was expected that the PV values would be lowest for oils in puddings treated with the highest concentration of extracts, but this was not the case. This might be because the antioxidant activity of chemical compounds was dependent on several factors. In addition to concentration, hydrophilic/hydrophobic balance, chemical structure (i.e. location and orientation of functional groups), temperature, reduction

Table 2. Peroxide, anisidine, thiobarbituric acid and iodine values of oils extracted from groundnuts pudding cooked with 0, 0.5, 1, 2 and 4% of *A. sativum* or *S. aromaticum* extract powders.

Peroxide values				
Samples	PV _{0.5%} (meqO ₂ /kg)	PV _{1%} (meqO ₂ /kg)	PV _{2%} (meqO ₂ /kg)	PV _{4%} (meqO ₂ /kg)
OAS	3.69 ± 0.76 ^{bb}	2.41 ± 0.35 ^{bb}	5.57 ± 0.32 ^{ba}	5.99 ± 0.45 ^{ba}
OZA	1.72 ± 0.00 ^{cc}	2.73 ± 0.73 ^{bc}	12.21 ± 0.89 ^{ca}	6.80 ± 1.74 ^{bb}
GPWS	18.39 ± 1.25 ^{aa}	18.39 ± 1.25 ^{aa}	18.39 ± 1.25 ^{aa}	18.39 ± 1.25 ^{aa}
Anisidine values				
	AnV _{0.5%}	AnV _{1%}	AnV _{2%}	AnV _{4%}
OAS	19.04 ± 0.72 ^{ba}	2.90 ± 0.57 ^{cc}	14.15 ± 4.02 ^{ba}	10.60 ± 1.43 ^{cb}
OZA	13.82 ± 3.15 ^{bc}	25.37 ± 0.00 ^{ba}	12.50 ± 0.23 ^{bc}	20.61 ± 0.38 ^{bb}
GPWS	49.49 ± 2.41 ^{aa}	49.49 ± 2.41 ^{aa}	49.49 ± 2.41 ^{aa}	49.49 ± 2.41 ^{aa}
Thiobarbituric acid values				
	TBA _{0.5%} (meqMDA/kg)	TBA _{1%} (meqMDA/kg)	TBA _{2%} (meqMDA/kg)	TBA _{4%} (meqMDA/kg)
OAS	6.31 ± 0.39 ^{ba}	5.21 ± 0.22 ^{bb}	7.56 ± 0.36 ^{aa}	5.91 ± 0.33 ^{bb}
OZA	5.65 ± 0.31 ^{bb}	4.19 ± 0.17 ^{bb}	7.00 ± 0.71 ^{aa}	5.10 ± 0.94 ^{bb}
GPWS	8.56 ± 0.16 ^{aa}	8.56 ± 0.16 ^{aa}	8.56 ± 0.16 ^{aa}	8.56 ± 0.16 ^{aa}
Iodine values				
	IV _{0.5%} (g I ₂ /100g)	IV _{1%} (g I ₂ /100g)	IV _{2%} (g I ₂ /100g)	IV _{4%} (g I ₂ /100g)
OAS	50.18 ± 0.00 ^{bb}	51.08 ± 1.24 ^{ab}	61.01 ± 2.54 ^{aa}	55.14 ± 1.06 ^{aa}
OZA	56.78 ± 0.44 ^{ab}	52.49 ± 2.44 ^{ac}	60.98 ± 0.79 ^{aa}	51.19 ± 0.20 ^{bc}
GPWS	49.00 ± 0.77 ^{ba}	49.00 ± 0.77 ^{ba}	49.00 ± 0.77 ^{ba}	49.00 ± 0.77 ^{ca}

Values with different superscripts in the same line in capital letters and in the same column in small letters differ significantly at $p < 0.05$.

OAS: Oil extracted from groundnuts pudding supplemented with *A. sativum* extract powder; **OZA:** Oil extracted from groundnuts pudding supplemented with *Z. aromaticum* extract Powder. **GPWS:** Oil extracted from groundnuts pudding without spice extract powder.

potential and composition of the antioxidant extract are known to alter the PV of foods (Esfandi et al., 2019; Kamal-Eldin and Appelqvist, 1996; Ratnasari et al., 2017). Indeed, at high concentrations, antioxidants can rather have prooxidative effects (Mariod et al., 2006). It will be important to determine the chemical composition of the extracts and, also to assay them at much lower concentrations than the ones used in the present work in order to determine the nature, the structure, the names of antioxidant present in our spice extract powders and the exact concentrations that can be used to prevent lipid oxidation of groundnuts pudding.

3.3.2. Anisidine value (AnV)

Despite the fact that hydroperoxides are very unstable under the effect of temperature, it would be advisable to test the secondary oxidation products before concluding on the oxidation state of oils extracted from groundnuts pudding after steam cooking (Eymard, 2003).

The effect of steam cooking on the anisidine value (AnV) of lipid extracted from groundnuts pudding cooked with four concentrations (0.5–4%) of *S. aromaticum*, or *A. sativum* extract powder is showed in Table 2. Oil extracted from groundnuts pudding cooked with 0.5, 1, 2, and 4%, of *S. aromaticum*, or *A. sativum* extract powder presented an AnV value significantly lower ($p < 0.05$) than that of the control groundnuts pudding cooked without spices. Oil extracted from groundnuts pudding cooked with 1% of *A. sativum* extract powder presented the lowest AnV and this value was significantly lower than oils extracted from groundnuts pudding cooked with *Allium sativum* powder. Concerning the anisidine values of oils extracted from groundnuts pudding cooked with *Syzygium aromaticum* extract powder, oil samples from groundnuts pudding with 2% inclusion of ZA extract powder presented an anisidine value significantly lower than those prepared with 1% and 4%. The high AnV of oil extracted from groundnuts pudding without added spice powder, may be the consequence of rapid decomposition of hydroperoxides and alkoxy radicals under the effect of temperature in favor of the secondary oxidation products (Eymard, 2003). However the low AnV obtained with OAS and OZA may be the consequence of antioxidants present in added spices that have the ability to inhibit free fatty acid form, thus preventing lipid oxidation (Womeni et al. (2013); Dudonne

et al. (2009)) since they possesses phenolic compounds and antioxidant properties such as DPPH, ORAC and FRAP. The lowest anisidine values observed with the mid-addition levels of extract powders may be the consequence of slow transformation of hydroperoxides in favor of secondary oxidation products. However, Yin et al. (2002) showed that *Allium sativum* was rich in selenium and organosulfides which are potent antioxidants capable of preventing lipid peroxidation.

3.3.3. Thiobarbituric acids value (TBARS)

The effect of steam cooking on the thiobarbituric acid (TBARS) level of oil extracted from groundnuts pudding cooked with *S. aromaticum* or *A. sativum* extract powders is showed in Table 2. At first glance oils extracted from groundnuts pudding cooked with 0.5, 1 and 4% presented TBARS values significantly lower ($p < 0.05$) than that of oil extracted from control groundnuts pudding. Oils from groundnuts pudding cooked with AS powders at inclusion rates of 1 and 4% presented the lowest TBARS value while for those cooked with *S. aromaticum* the lowest value was observed with extract powder inclusion rates of 1 and 4%. The low TBARS observed compared to groundnuts pudding without added spice extract powder may be due to the effect of phenolic or flavonoids compounds found in these spices which would have prevented the conversion of the hydroperoxides into malondialdehyde and preserve the quality of lipid (Roman et al. (2015); Womeni et al. (2013)). However, the high values of TBARS for oils from samples treated with AS extract powders at inclusion rates of 0.5 and 2% and for ZA at 2% might be the consequence of low or high concentrations of antioxidant level. At high concentrations, antioxidants can rather have prooxidative effects (Mariod et al., 2006).

3.3.4. Iodine value (IV)

Table 2 shows the effect of *S. aromaticum* or *A. sativum* powder on the IV of oils extracted from groundnuts pudding after steam cooking. It can be observed that all groundnuts pudding cooked with spice extracts with the exception of that cooked with 0.5% of *A. sativum* presented IV significantly higher ($p < 0.05$) than that of oil extracted from pudding cooked without spice extract powder. Oils from groundnuts pudding treated with AS extract powders at inclusion rates of 2 and 4% and that of ZA extract powders at inclusion rates of 2% presented IV higher than

those of all oils extracted from pudding cooked with *A. sativum* or *S. aromaticum* respectively. The decreased in IV for oil from groundnuts pudding without added spice extract powder may be due to loss of double bonds of fatty acids due to oxidative loss during cooking (Womeni et al., 2013).

3.3.5. Correlation between the phenolic content (TPC), flavonoid content (FC), free radical scavenging (DPPH), ferric reducing antioxidant power (FRAP), hydroxyl radical (ROH), oxygen radical absorbance capacity (ORAC), peroxide value (PV), p-anisidine value (AnV), thiobarbituric acid value (TBARS) and iodine value (IV)

Pearson's correlation between the phenolic content (TPC), flavonoid content (FC), free radical scavenging (DPPH), hydroxyl radical (ROH), Oxygen radical absorbance capacity (ORAC), peroxide value (PV), p-anisidine value (AnV), thiobarbituric acid value (TBARS) and iodine value (IV) is presented in Table 3. There was a significant positive correlation at $p < 0.01$ ($r = 1.000$) between TPC and FC. A significant positive correlation at $p < 0.01$ ($r = 1.000$) between DPPH, TPC and FC ($r = 1.000$). Also, a significant positive correlation at $p < 0.01$ ($r = 1.000$) was observed between ORAC, TPC, FC, DPPH, ROH and FRAC. However, a significant negative correlation at $p < 0.01$ ($r = -1.000$) existed between IP, TPC, FC, DPPH, ROH, FRAP and ORAC. It also appears that, AnV was significantly negative correlated at $p < 0.01$ ($r = -1.000$) to TPC, FC, DPPH, ROH, FRAP, ORAC and positively correlated at $p < 0.01$ ($r = 1.000$) to PV. The IV was positively correlated to TPC, FC, DPPH, ROH, FRAP, ORAC and negatively and significantly correlated to PV, AnV, and TBARS at $p < 0.01$ ($r = -1.000$). Regarding the significant positive correlation observed between the phytochemical and the antioxidant properties of these extract powder, there exists a relation between the TPC, FC and DPPH free radical scavenging because the extract powder that presented a high antiradical activity with regards to DPPH also has the highest phenolic and flavonoid contents. The significant positive correlation observed between PV, AnV and TBARS may be the consequence of rapid transformation of primary products to secondary oxidation products since hydroperoxides are very unstable under the effect of temperature (Eymard, 2003). Results showing the negative and significant correlation between IV, PV, AnV and TBARS are in line with those of Hamid (2011) who showed that IV of oils extracted from chicken, cattle and camel meat during refrigerated storage is negatively correlated to PV and TBARS.

3.3.6. Fourier transform infrared spectrometry (FTIR)

Oils extracted from groundnuts pudding cooked with 0.5% of *A. sativum* and 0.5% of *S. aromaticum* extract powders was used for Fourier transform infrared spectrometry in order to confirm thier

oxidation state. Fourier transform infrared spectroscopy (FTIR) makes it possible to qualitatively determine the organic components of a sample since the mode of vibration of each group causes the appearance of peaks at a specific frequency. Figure 2 shows the spectra of oils extracted from groundnuts pudding cooked with 0.5% AS extract powder (a), 0.5% ZA extract powder (b) and that of pudding cooked without spice extract powder (c). Peaks appearing on these spectra at 3453.4; 3476.7 and 3470.8 cm^{-1} could be associated with O–H stretching vibration of hydroperoxides as the oxidation of polyunsaturated lipids underwent peroxidation reactions with oxygen and formed lipid hydroperoxides. Lu et al. (2014) in their work on the characterization of rapeseed oil using FTIR-ATR revealed the appearance of this band at 3500 cm^{-1} . The band observed at 3007 cm^{-1} is assigned to C–H stretching vibration of the cis double bond (=CH). Similar result was obtained by Chauhan et al. (2016) at 3007 cm^{-1} . This band was also observed by Lu et al. (2014) and Vlachos et al. (2006) respectively at 3010 cm^{-1} and at 3006 cm^{-1} . Peaks that appear around 2927 and 2854 cm^{-1} were assigned to symmetric and asymmetric stretching vibration of the aliphatic CH_2 group. Shayla et al. (2018) observed these bands at 2921 cm^{-1} . This peak was also observed by Lu et al. (2014) at 2924 cm^{-1} . The strong bands observed at about 1747 cm^{-1} is associated to the ester carbonyl functional group of the triglycerides (C=O) in the oil samples. Similar results were observed by Shayla et al. (2018) and Lu et al. (2014) at 1745 and 1746 cm^{-1} respectively. In addition, Lei et al. (2017) also observed these bands at 1745 cm^{-1} during the analysis of vegetable oils. The band observed around 1653 cm^{-1} was associated to C=C stretching vibration of cis-olefins. This band was also observed by Lu et al. (2014) at 1650 cm^{-1} . The bands obtained around 1376 and 1463 cm^{-1} were characteristic of bending vibrations of the CH_2 and CH_3 aliphatic groups. These results are similar to those observed by Lei et al. (2017) at 1461 and 1376 cm^{-1} . The absorption spectra that appear between 1238 and 700 cm^{-1} , is generally called fingerprints. When present, it makes it possible to precisely characterize molecules. The peak observed between 1238 and 1163 cm^{-1} was the Stretching vibration of the C–O ester groups. These bands were observed by Lu et al. (2014) at 1240 and 1160 cm^{-1} . The peak observed at 722 cm^{-1} was assigned to overlapping of the CH_2 rocking vibration and the out-of-plane vibration of cis disubstituted olefins. Lei et al. (2017) and Chauhan et al. (2016) also observed these bands 723 and 719 cm^{-1} respectively. The critical absorption bands associated with common oxidation end products of lipid oxidation during treatment could be observed in region 3800–3200 cm^{-1} . This is the –OH stretching region (Bhundit et al., 2004). However, the band at 3470 cm^{-1} has been reported to be associated with the –OH stretching frequency of hydro peroxide. Relating the absorption frequency 3470.8 cm^{-1} with the PV, AnV, TBA value and the IV we can observe that oil extracted from groundnuts pudding cooked without spice extract powder present PV and

Table 3. Correlation between phenolic content (TPC), flavonoid content (FC), free radical scavenging (DPPH), ferric reducing antioxidant power (FRAP), hydroxyl radical (ROH), Oxygen radical absorbance capacity (ORAC), peroxide value (PV), p-anisidine value (AnV), thiobarbituric acid value (TBARS) and iodine value (IV).

	TPC	FC	DPPH	ROH	FRAP	ORAC	PV	AnV	TBARS	IV
TPC	1									
FC	1.000**	1								
DPPH	1.000**	1.000**	1							
ROH	1.000**	1.000**	1.000**	1						
FRAP	1.000**	1.000**	1.000**	1.000**	1					
ORAC	1.000**	1.000**	1.000**	1.000**	1.000**	1				
IP	-1.000**	-1.000**	-1.000**	-1.000**	-1.000**	-1.000**	1			
AnV	-1.000**	-1.000**	-1.000**	-1.000**	-1.000**	-1.000**	1.000**	1		
TBARS	-1.000**	-1.000**	-1.000**	-1.000**	-1.000**	-1.000**	1.000**	1.000**	1	
IV	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	-1.000**	-1.000**	-1.000**	1

** Correlation differ significantly at 0.01.

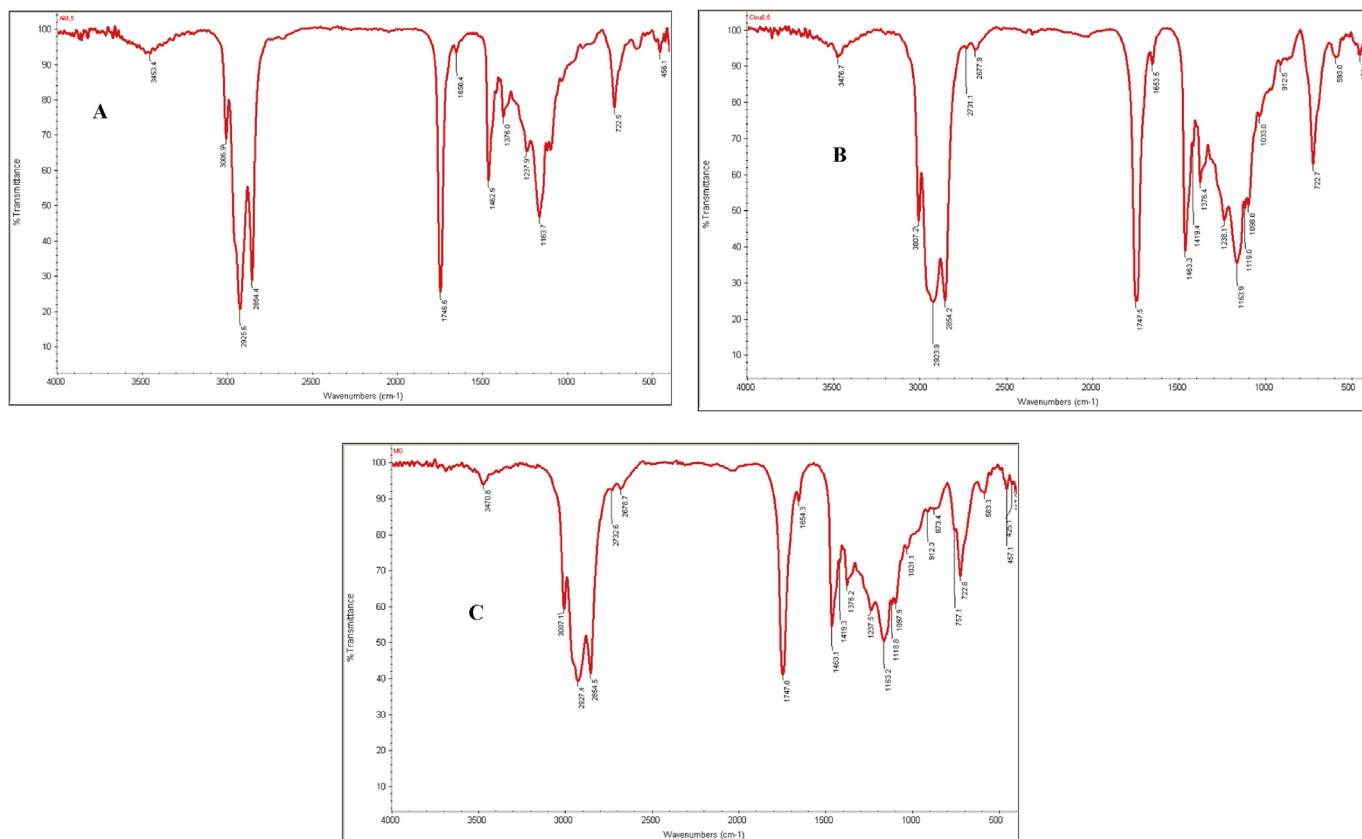


Figure 2. Fourier transform infrared (FTIR) spectra of oil extracted from groundnuts pudding cooked with (A) 0.5% of *A. sativum*; (B) 0.5% of *S. aromaticum*; and (C) without spice extract powder.

TBA higher than those of oils extracted from pudding cooked with 0.5% of *A. sativum* and 0.5% of *S. aromaticum* extract powders. The IV of pudding cooked without spice extract powder was lower than that of pudding cooked with 0.5% AS extract powder and 0.5% ZA extract powder. Regards to this it is clear that oil extracted from pudding cooked without spice extract powder was deteriorated and this is supported by FTIR.

4. Conclusion

The effect of *S. aromaticum* or *A. sativum* spice extract powders added at different concentrations in groundnuts paste for the preparation of groundnuts pudding during steam cooking revealed that these spices limit the formation of primary, secondary oxidation products and alteration of double bonds of groundnuts pudding oil since they possess phytochemical and antioxidant activity. Regards to these observations these spices can be used to prevent lipid oxidation during cooking of oily foods.

Declarations

Author contribution statement

Hermann A. K. Foffe: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Serge C. N. Houketchang, Fabrice T. Djikeng, Gires B. Teboukeu: Performed the experiments; Analyzed and interpreted the data.

Apollinaire Tsopmo, Hilaire M. Womeni: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data.

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The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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