

Comparison of Antioxidant Activity of Rosemary Extract in Peanut Oil and Flaxseed Oil in Relation to their Fatty Acids Compositions

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Abstract: In this investigation, the antioxidant activity of rosemary extract in peanut oil (PO) and flaxseed oil (FO) during accelerated storage has been studied and compared. Rosemary extracts were prepared in two forms, *i.e.* powdery rosemary extract (PRE) and oily rosemary extract (ORE). The changes of fatty acids composition during 24 days of storage were measured, and the correlations between unsaturated fatty acids (18:3, 18:2 and 18:1) and parameters of oil oxidation (PV, *p*-AV and TBARS) were analyzed, respectively. PRE was more effective than ORE in stabilization of both PO and FO samples. PO samples showed significantly higher oxidative stability than FO samples during 24 days of storage. Negative correlations were found in 18:3 vs. peroxide value in PO and FO samples. Positive correlations were found in 18:1 vs. *p*-AV and TBARS in PO and FO samples, and in 18:2 vs. *p*-AV and TBARS only in FO samples. Hence FO samples were more susceptible to oxidation than PO samples because of its significantly higher content of 18:3. Moreover, rosemary extract effectively inhibited the oil oxidation potentially by retarding the decomposition of 18:1 for PO samples, while by retarding the decomposition of 18:2 and 18:1 for FO samples.

Practical applications: Oxidation of unsaturated fatty acids is one of the major concerns in oil consumption and storage. The addition of natural antioxidants is a potential way to retard the oxidation of oils and thus to extend the shelf life of oil products. Rosemary extract has been reported to show good antioxidant activity and could act as a good resource to inhibit oil oxidation. The results in this study not only confirmed the effectiveness of rosemary extract in stabilizing oil during storage, but also suggested different anti-oxidative pathways in different oil system in relation to their different fatty acid compositions. Therefore, rosemary extract may have great market potential as food antioxidant in edible oils.

Keywords: Antioxidant activity, Peanut oil, Flaxseed oil, Fatty acid composition, Rosemary extract.

1. INTRODUCTION

Peanut oil is an essential component of the Chinese diet, consumption of peanut oil may improve lipid profiles without promoting weight gain because of its high-unsaturated fat content [1]. The interest in the consumption of flaxseed oil is related to its high content (50-55% total fatty acids) of linolenic acid [2], which has potential to reduce the risk factors for cancer and cardiovascular disease [3]. However, one of the major concerns on consumption of vegetable oils is the oxidation of unsaturated fatty acids during storage, which would affect the shelf life of products. The oxidative stability of vegetable oils is influenced by illumination, availability of oxygen, temperature, fatty acid composition, and nature and concentration of antioxidants [4]. Differences in fatty acids composition are translated to differences in the oxidative stability of different oils [5], thus the oxidative stability of peanut oil and flaxseed oil may be related to their different fatty acid profiles.

Previous studies indicated that addition of antioxidants could prevent or retard the oxidation of oils and thus could extend the shelf life of oil products [4-16, 20, 22, 25]. Some synthetic antioxidants such as

butylatedhydroxyanisole (BHA), butylatedhydroxytoluene (BHT) and *tert*-butylhydroquinone (TBHQ) have been used as additive to retard lipid oxidation in food [6]. However, the application of these synthetic antioxidants are restricted by their suspected toxic properties. Therefore, search for natural antioxidants with less toxicity is now a trend in both research and industrial applications [7]. Actually, several plant materials containing phenolic compounds such as grape seed, black pepper and rosemary and oriental herbs have been reported to be effective in retarding lipid oxidation [8-11]. The effect of rosemary extract on decreasing oil oxidation has been investigated in many aspects [10, 13, 14, 16]. However, studies of its antioxidative effectiveness in relation to the oil fatty acids composition have been rarely reported.

Therefore, the objective of this work was to compare the antioxidant activity of rosemary extract in peanut oil (rich in linoleic acid, 18:2) and flaxseed oil (rich in linolenic acid, 18:3). The oxidative stability was evaluated by peroxide value (PV), acid value (AV), *p*-anisidine value (*p*-AV) and thiobarbituric acid-relative substances assay (TBARS). Moreover, the relations between the changes of oxidative stability and the changes of fatty acids composition (18:3, 18:2 and 18:1) were analyzed. The possible antioxidative mechanisms of rosemary extract in peanut oil and in flaxseed oil under accelerated storage were discussed, respectively.

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2. MATERIALS AND METHODS

2.1. Materials and Reagents

Refined, bleached and deodorized (RBD) peanut oil (PO) was purchased from a local market. RBD flaxseed oil (FO) was provided by Hebei Gunn Reiss Biotechnology Co., Ltd. (Zhangjiakou, Hebei, China). Powdery rosemary extract (PRE) and oily rosemary extract (ORE) were of food grade and provided by Guizhou Red Star Development Co., Ltd. (Duyun, Guizhou, China). All the chemicals and reagents used were of analytical reagent grade and were purchased from China Pharmaceutical Group Chemical Reagent Co., Ltd. (Beijing, China). Fatty acids methyl ester (FAMES) (C14:0, C16:0, C16:1*n* - 7, C17:0, C18:0, C18:1*n* - 9, C18:1*n* - 7; C18:2*n* - 6, C18:3*n* - 3, C20:0, C20:1*n* - 11, C22:0, C22:1*n* - 9, C22:2*n* - 6, C24:0), squalene and squalane were from Sigma Chemical Co (St. Louis, MO, USA).

2.2. Sample Preparation

Powdery rosemary extract (PRE) and oily rosemary extract (ORE) were added to preheated RBD peanut oil (PO) and flaxseed oil (FO) (at 50°C for 3 h) at concentrations of 0.07%, 0.07%, and 0.04%, respectively. All the samples (150 mL each) were placed in dark brown colored reagent bottles with narrow necks, without stoppers and stored in an oven at fixed temperature of 60°C. Control samples were also placed under the same storage conditions. Analyses were carried out after regular intervals of 4 days (96 h). At least three samples of each category were analyzed to fulfill the requirements for statistical analyses.

2.3. Measurement of Peroxide Value (PV)

The peroxides are intermediate products in the formation of carbonyl and hydroxyl compounds. The primary products of lipid oxidation are hydroperoxides, which are generally referred to as peroxides; therefore, it is reasonable to determine the concentration of peroxides as a measure of the extent of oxidation. Peroxide value was determined according to AOAC method 965.33 with some modifications [17], and expressed as meq O₂/kg of lipid. Standard deviation for each PV determination was less than 2%. 50 mL of acetic acid-isooctane (3:2, v/v) solution was added to 2 g aliquots of the oil-antioxidant mixtures, 0.5 mL saturated KI aqueous solution was added. The mixture was shaken by hand for 1 min and kept in the dark for 1 min. After the addition of 30 mL distilled water, the

mixture was titrated against sodium thiosulfate (0.01 M) until the yellow color almost disappeared. Then 1.0 mL starch indicator solution was added. Titration was continued until the blue color by the starch indicator has just disappeared. The blank was analyzed under similar conditions. The PV is calculated by the following equation.

$$PV \text{ (meq/kg)} = 1000 \times c \times (V - V_0) / m$$

where *c* is the exact normality of sodium thiosulfate solution used for titration; *V* is titre of 0.01 M sodium thiosulfate solution (mL); *V*₀ is the blank titre (mL); *m* is the weight in grams of sample used.

2.4. Measurement of Acid Value (AV)

Acid value (AV), a measure of free fatty acids from hydrolysis of triglyceride, is usually considered to be one of the main parameters to reflect the quality of oils and the quality change during storage. Acid values were determined by a standard titration method according to AOCS method Cd 3d-63[18]. Briefly, 5g sample was completely dissolved in 50 mL of previously neutralized diethyl ether-ethanol medium (2:1, v/v) solution. Using an ethanolic solution of 0.05 M potassium hydroxide as the standard reagent to a phenolphthalein endpoint (the pink color of the phenolphthalein persisted for at least 30 s). AV value was expressed as milligrams of potassium hydroxide required to neutralize the free fatty acids present in 1 g of oil sample (mg KOH/g). The AV was calculated as:

$$AV \text{ (mg/g)} = 56.1 \times V \times c / m$$

where *V* is the volume in milliliters of standard volumetric potassium hydroxide solution used; *c* is the concentration in moles per liter of the standard volumetric potassium hydroxide solution used; *m* is the mass in grams of the test sample of oil samples.

2.5. Measurement of *p*-Anisidine Value (*p*-AV)

p-anisidine value is a measurement of aldehyde content in oil, principally 2,4-dienals and 2-alkenals, which may contribute to off-flavor of oxidized oils. Aldehydes are secondary oxidation products produced during the oxidation of lipids. *p*-anisidine value was determined according to AOCS Official Method Cd 18-90 [19]. Standard deviation for each *p*-anisidine value (*p*-AV) determination was less than 0.1. Briefly, 0.4-8 g aliquots of oil-antioxidant mixtures were dissolved and diluted to volume (25 mL) with iso-octane and the absorbance read at 350 nm (*A*_b). 1.0 mL of *p*-anisidine

reagent was added to 5 mL of diluted sample. A reference solution was also made up with 5.0 mL of iso-octane and 1.0 mL of *p*-anisidine reagent. Solutions were left to stand for 10 min and the absorbance read again (A_s). The *p*-AV was calculated by the following equation:

$$p\text{-AV} = 25 \times (1.2A_s - A_b)/m$$

where A_s is the absorbance of the oil solution after reaction with the *p*-anisidine reagent; A_b is the absorbance of the oil solution; m is the mass in grams of the initial test portion diluted with isooctane.

2.6. Measurement of Thiobarbituric Acid-Reactive Substances Assay (TBARS)

TBARS measures the formation of secondary oxidation products *i.e.* pentanal, hexanal, 4-hydroxynonal and malondialdehyde (MDA). TBARS was measured by the reaction of these products with TBA. Lipid oxidation of all samples was assessed by the 2-thiobarbituric acid (TBA) method according to GB/T 5009.181-2003 and method with some modifications [20]. 5 g of samples were homogenized in 10 mL of TCA (7.5%)-EDTA (0.1%) solution. The samples was shaken continuously for 0.5 h with a mechanical shaker with the rotation speed of 150 r/min at room temperature, and then filtered. Exactly 3 mL of filtrate was added to 3 mL of TBA solution in a 10 mL colorimetric tube, and heated in a water bath for 40 min. The mixed solution was allowed to stand for 1 h. The absorbance was measured at 532 nm UV spectrophotometer UV-1800 (Shimadzu Corporation, Japan). TBARS were calculated from a standard curve of malondialdehyde (MDA), freshly prepared by acidification of TEP (1,1,3,3-tetraethoxypropane) in the range from 0.02 to 0.3 $\mu\text{g/mL}$ ($y = 0.2622x + 0.0013$,

$R^2 = 0.9999$), and expressed as mg of malondialdehyde per kg oil.

2.7. Analysis of Fatty Acids Composition

The fatty acids composition of the oils was analyzed by gas chromatography (GC) throughout the experiments. The preparation of fatty acid methyl ester (FAME) and gas chromatographic analysis were conducted according to the method [21] with some modifications.

Preparation of fatty acid methyl ester (FAME): 1 g each of peanut-antioxidant and flaxseed-antioxidant oil samples was methylated in screw capped glass tubes. Derivatization was performed with 15 mL of methanolic acetyl-chloride (10%, v/v) at 80°C to 10 mL tubes and centrifuged at 5000g for 5 min. Supernatant was then transformed to previously dried auto-sampler vial and waited for GC analysis.

Gas chromatographic analysis: A gas chromatograph 7890A-5975C (Agilent Technologies, Inc. USA) equipped to 10 mL tubes and centrifuged at 5000g for 5 min. Supernatant was then transformed to previously dried auto-sampler vial and waited for GC analysis.

Gas chromatographic analysis: A gas chromatograph 7890A-5975C (Agilent Technologies, Inc. USA) equipped with a FID detector, an auto sampler, a split-splitless injector, and an integrator was used for FAMES analysis. The capillary column was a fused silica DB-5 (30 m \times 0.25 mm i.d.; 0.25 μm film thickness; Agilent Technologies, Inc. USA) under the following temperature program: the initial oven temperature 180°C was hold for 10min, and then programmed to increase with 15°C/min to 210°C, with 3°C/min to 230°C, to reach finally with 5°C/min 280°C and hold for 2min. Each 1.0 μL of samples were

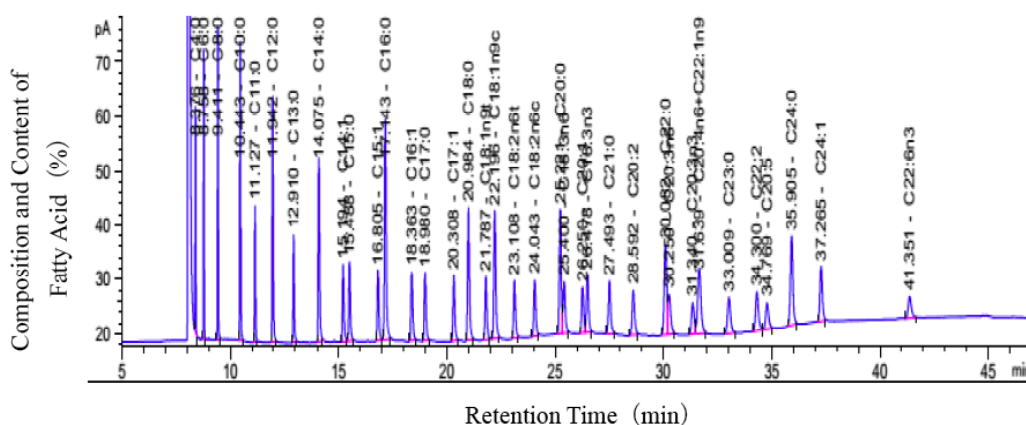


Figure 1: Composition and content of 37 kinds of fatty acid methyl ester standards.

introduced to the column via a split injector (split ratio 1:50) at 250°C and the flow rate of nitrogen, used as carrier gas, was 1.2mL/min. Temperature of both split injector and flame ionization detector was 280 °C. Standard FAMES stock solutions were prepared in hexane and for identification of fatty acid composition in peanut oils and in flaxseed oils. Data acquisition and processing were performed with the GC-S Solution Software (Agilent Technologies, Inc. USA). The percentage composition of the oils was calculated from GC peak areas without using correction factors. A chromatogram represents composition and content of 37 kinds of fatty acid methyl ester standards in Figure 1. A chromatogram represents composition and content of ORE-stabilized samples for flaxseed oil in Figure 2.

2.8. The Nomenclature Used in the Paper

Table 1 indicates the nomenclature used and the meaning for better understanding of the text.

Table 1: The Acronyms Represent the Meaning in the Paper

Acronym	Full Name
PO	Peanut Oil
FO	Flaxseed Oil
RBD	Refined, bleached and deodorized
PRE	Powdery Rosemary Extract
ORE	Oily Rosemary Extract
PV	Peroxide Value
AV	Acid Value
P-AV	P-anisidine Value
TBARS	Thiobarbituric Acid-relative Substances
GC	Gas Chromatography

2.9. Statistical Analysis

Analysis of variance (ANOVA) was carried out by Microcal Origin 8.5 (Microcal Software, Inc., Northampton, MA, USA) and by SPSS Statistics 17.0 (SPSS Inc., Chicago, IL, USA). ANOVA tests were performed to determine the significance at 95% confidence. Each experiment was performed in triplicates.

3 RESULTS

3.1. Effect on Peroxide Value

The relative increase of peroxide value (PV) for peanut oil samples and flaxseed oil samples are shown in Figure 3. A continuous increase in PV with the increase of storage time was observed for PO samples, and a common trend for FO samples. The increase of PV was due to the formation and accumulation of hydroperoxides *i.e.*, primary oxidation products. For both PO and FO samples, the highest PV was observed for control samples followed by ORE-stabilized samples and PRE-stabilized samples, which indicated the higher efficiency of PRE than ORE in retarding the oil oxidation. However, no significant differences of PV was found between PRE-stabilized samples and ORE-stabilized samples for FO in the 24 days of storage and for PO in the initial 20 days of storage ($p>0.05$), whereas significant differences was found in the 24th day of storage for PO samples stabilized with PRE and ORE ($p<0.05$). For PO samples, the increase in PV was very low initially and the difference in PV of control and stabilized oil samples were not noticeable, but it increased significantly near 4th day of storage and went on increasing further with the increase of storage time,

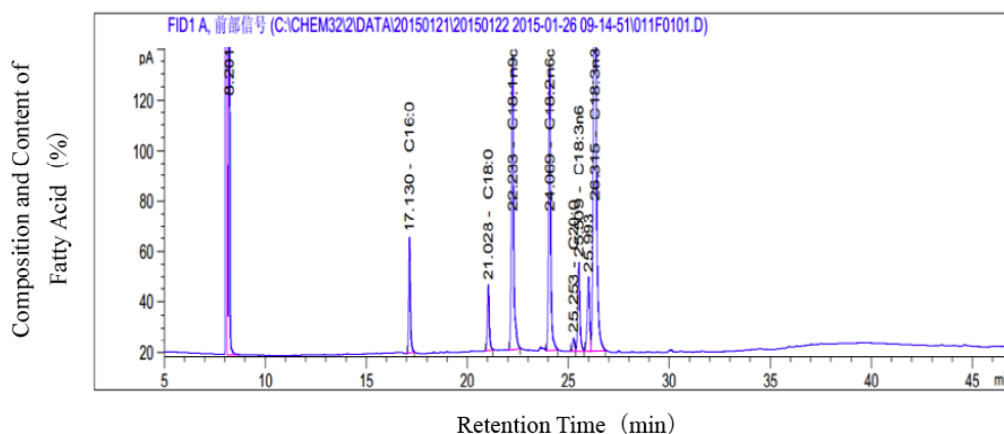


Figure 2: Composition and content of ORE-stabilized samples for flaxseed oil.

reaching the maximum values after 24 day of storage. However, the increase in PV for FO samples was very fast in the 1st day of storage, but it was significantly higher in the control than in the stabilized samples ($p < 0.05$). The lower increase for the stabilized samples was attributed to the interruption of oil deterioration by antioxidants in the early stages, which delayed the onset of the reaction.

After storage upto 24 days, PVs were 4.01 and 5.67 meq/kg for PO samples stabilized with PRE and ORE, and maximum value was from the control of PO (6.53 meq/kg). PV increased 3.73 fold for the control of PO while it increased 1.90 and 3.10 fold for PO samples stabilized with PRE and ORE. In comparison, maximum of PV reached its highest value from the control (6.17 meq/kg) of FO and increased 25.80 fold of its initial peroxide value. Furthermore, PV was 4.66 and 4.63 meq/kg for FO samples stabilized with PRE and ORE, which increased 19.26 fold and 19.13 fold in the 24th day of storage. Apparently, the increases of PV in FO samples were significantly higher than that in PO samples ($p < 0.05$). It was indicated that the oxidation rate of flaxseed oil was higher than that of peanut oil, which might be attributed to their different fatty acids composition. Previous studies showed that flaxseed oil was susceptible to oxidation because of its high content of linolenic acid.

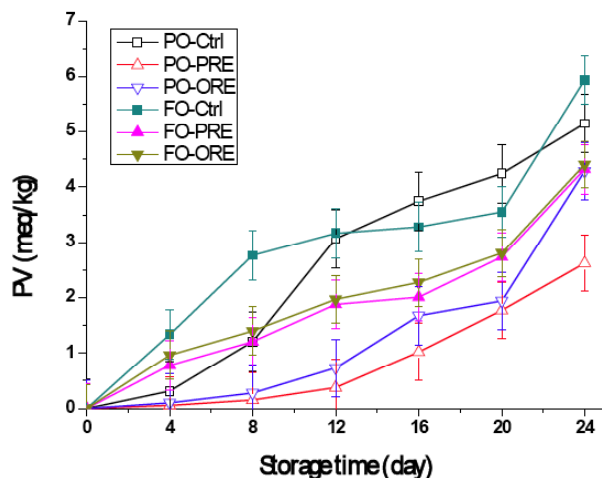


Figure 3: Relative increase in peroxide value (PV) of peanut oil and flaxseed oil samples under accelerated storage. PO-Ctrl represents the control samples for peanut oil, PO-PRE represents powdery rosemary extract stabilized samples for peanut oil, PO-ORE represents oily rosemary extract for peanut oil, FO-Ctrl represents the control samples for flaxseed oil, FO-PRE represents powdery rosemary extract stabilized samples for flaxseed oil, FO-ORE represents oily rosemary extract stabilized samples for flaxseed oil, the same below.

3.2. Effect on Acid Value

The relative increase of acid value (AV) for peanut oil samples and flaxseed oil samples are presented in Figure 4. Similar to the results of peroxide values, Lowest PV was observed for samples stabilized with PRE in each interval, which indicated that PRE was more effective in both peanut oil and flaxseed oil. However, no significant differences were found between PRE-stabilized samples and ORE-stabilized samples for both FO and PO ($p > 0.05$). The increase of AV as function of storage time was observed for all samples. It increased significantly from the very beginning and went on increasing much slower after 4 days of storage. The increase of AV in the control of FO was significantly higher than that in the control of PO in 24 days of storage ($p < 0.05$). A pronounced increase of AV was observed from the control of FO in the 16th day of storage. Significant differences were observed between the stabilized samples and the control for both PO and FO ($p < 0.05$). The initial acid value was 0.93 mg KOH/g for PO samples and 0.40 mg KOH/g for FO samples. After storage up to 24 days, AV increased 0.18 fold of the initial value for the control of PO samples (0.17 mg KOH/g) and it increased 0.04 and 0.07 fold for PO samples stabilized with PRE (0.03 mg KOH/g) and ORE (0.06 mg KOH/g), respectively. However, much higher increases were found in FO samples in comparison. AV increased 1.06 fold for the control (0.43 mg KOH/g) and it increased 0.66 and 0.69 fold for samples stabilized with PRE (0.27 mg KOH/g) and ORE (0.28 mg KOH/g).

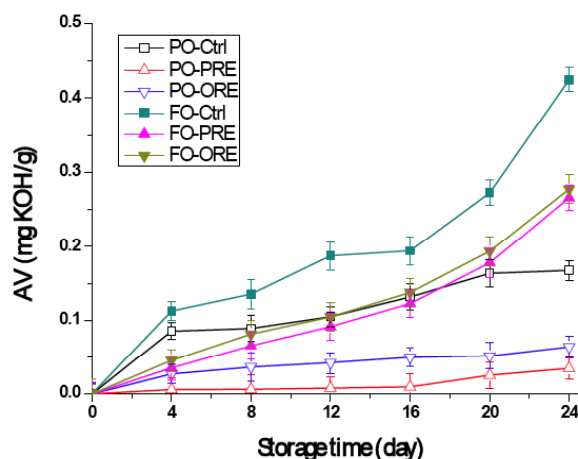


Figure 4: Relative increase in acid value (AV) of peanut oil and flaxseed oil samples under accelerated storage.

Generally, AV increases as a result of the formation of fatty acids by hydrolysis and by oxidative reactions. The study that investigated the antioxidant efficacy of

methanolic extracts of peanut hulls in peanut oils showed that the AV of peanut oils with antioxidants was significantly ($p < 0.05$) lower than that of the control, and it rapidly increased at 35 day of storage [22]. Though significant increase of AV was not observed throughout 24 day of storage in peanut oils, the rapidly increase in flaxseed oil without antioxidants at 16th day of storage may indicate decomposition of peroxides. Referring to our results of PV, it was indicated that antioxidants not only prevented the formation of peroxides but also inhibited its decomposition in flaxseed oil and further increased the oxidative stability of flaxseed oil.

3.3. Effect on *p*-Anisidine Value

The increases of *p*-anisidine (*p*-AV) are shown in Figure 5. A continuous increase of *p*-AV was observed for both PO and FO samples in the 24 day of storage. PO without antioxidants reached its maximum of 2.86 in the 24th day of storage with a significant increase in the 12th day of storage. FO without antioxidants reached its maximum of 10.50 in the 24th day of storage with a significant increase in the initial 4 days of storage, and went on increasing with lower level of average increase each interval afterwards. The significant higher increase of *p*-AV in control of FO (2.66 fold) than that of PO (1.71 fold) may be attributed to the higher oxidative susceptibility of flaxseed. A significant difference in *p*-AV was found in FO between the control and the stabilized samples ($p < 0.05$). However, no significant difference in *p*-AV was found in PO between the control and the stabilized samples ($p > 0.05$). It was showed the antioxidant activities of 0.07% PRE was significantly more effective ($p < 0.05$) than that of 0.07% ORE in the initial 12 days of storage for FO samples, while no significant differences were found in PO samples even though the lowest *p*-AV was obtained in samples containing PRE. Furthermore, the superiority of PRE in decreasing the *p*-AV of both PO and FO was in agreement with our previous results of PV and AV.

The degradation of the primary oxidation products to secondary oxidation products with aldehydic and ketonic functions negatively affects their flavor [5]. The delay in the formation of aldehydes by chemical methods can be used to evaluate the efficacy of antioxidants. The oxidative effectiveness of PRE and ORE in decreasing the *p*-anisidine value may be attributed to their ability to slow the rate of aldehydes formation. A study reported that good quality oil should

have a *p*-anisidine value of less than two [23]. Another investigated the quality of cold-pressed flaxseed oils and obtained significant lower *p*-anisidine value (varied between 0.36 and 0.74) than ours, it was inferred that processing method, especially temperature, significantly influenced the oxidative stability [24].

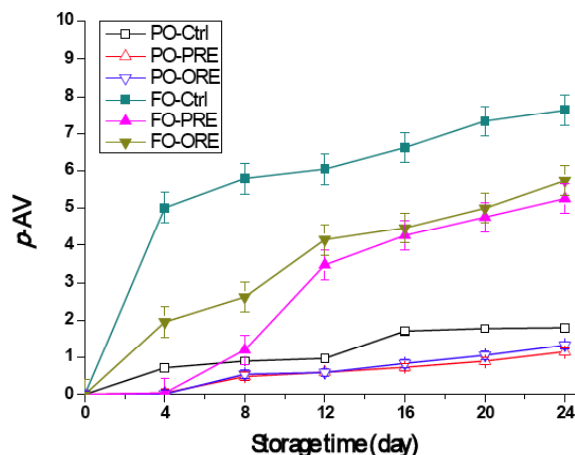


Figure 5: Relative increase in *p*-anisidine value (*p*-AV) of peanut oil and flaxseed oil samples under accelerated storage.

3.4. Effect on Thiobarbituric Acid-Relative Substances (TBARS)

Increases of TBARS up to 24 days of storage for control and stabilized samples are shown in Figure 6. The same order of antioxidant efficiency in TBARS was observed as was observed in the case of other assays *i.e.*, powdery rosemary extract > oily rosemary extract. A continuous increase in TBARS was observed for all the samples with the increase in storage period. Control exhibited the highest TBARS at all the stages

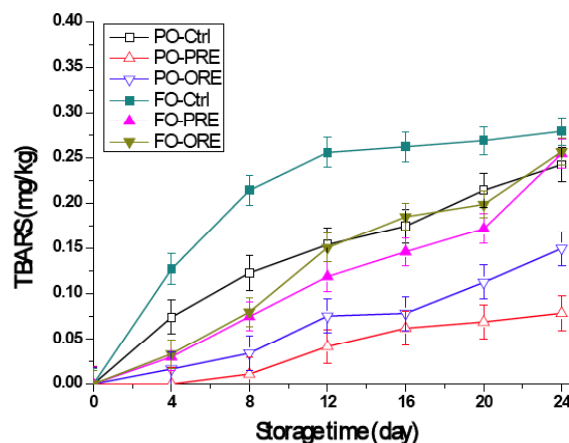


Figure 6: Relative increase in thiobarbituric acid-relative substances (TBARS) of peanut oil and flaxseed oil samples under accelerated storage.

of analysis during storage for both PO and FO samples. However, the increase of TBARS for FO samples slowed in the 12th day of storage for both control and stabilized flaxseed oils. After 24 days of storage, TBARS were in the range of 0.10-0.17 mg/kg peanut oil for stabilized samples with significant differences ($p < 0.05$) and 0.26 mg/kg peanut oil for its control sample. Meanwhile, TBARS was 0.28 mg/kg flaxseed oil for both PRE-stabilized samples and ORE-stabilized samples, and 0.30 mg/kg flaxseed oil for its control sample. Furthermore, PRE was more effective than ORE for both peanut and flaxseed oil. In the storage of 24 days, significant differences in TBARS was found between the control and PRE-stabilized sample in peanut oil in each interval ($p < 0.05$). However, no significant difference was found between the control and flaxseed oil containing this antioxidant

in the 24th day of storage ($p > 0.05$). It may be hypothesized that polyphenol antioxidants inhibit lipid peroxidation at the cost of their own life. Previous studies reported that only those antioxidants are to be preferred which have good effectiveness over longer periods and drastic conditions [25]. So it was inferred that the PRE was more effective in peanut oil than in flaxseed oil over 24 days of storage.

3.5. Effect on Fatty Acids Composition

Relative percentages of unsaturated fatty acids (18:3, 18:2 and 18:1) in peanut and flaxseed oil samples in the 24 days of storage are shown in Table 2. Linoleic acid (18:2) was chosen as an index to analyze peanut oil as it constituted large amount (about 35.85%) of its initial unsaturated fatty acids. The

Table 2: Relative Percentages of Unsaturated Fatty Acids (18:3, 18:2 and 18:1) in Peanut Oil and Flaxseed Oil Samples under Accelerated Storage

	Time (day)	Peanut Oil			Flaxseed Oil		
		C18:3 (%)	C18:2 (%)	C18:1 (%)	C18:3 (%)	C18:2 (%)	C18:1 (%)
Control	0	1.2251	35.8483	39.9908	50.0521	16.3894	15.6848
	4	1.1676	34.8065	39.8472	49.7882	16.3886	15.6816
	8	1.1589	34.6521	39.8076	49.7850	16.3710	15.6777
	12	1.1548	34.4879	39.8044	49.6336	16.3597	15.6689
	16	1.1402	34.2117	39.7814	49.5173	16.3555	15.6641
	20	1.1342	34.1906	39.7387	49.2573	16.3408	15.6486
	24	1.1201	33.9594	39.6840	49.0751	16.3170	15.5327
Rosemary (powdery)	0	1.2251	35.8483	39.9908	50.0521	16.3894	15.6848
	4	1.2239	35.8013	41.3975	49.9878	16.4234	15.7057
	8	1.1929	35.6620	41.4062	49.9299	16.4234	15.7488
	12	1.1605	35.4978	41.5387	49.7514	16.4255	15.7762
	16	1.1568	35.4374	41.6747	49.7257	16.4336	15.7849
	20	1.1462	35.3984	41.6927	49.6890	16.4893	15.8557
	24	1.1345	35.3623	41.7956	49.4949	16.5386	16.0593
Rosemary (oily)	0	1.2251	35.8483	39.9908	50.0521	16.3894	15.6848
	4	1.2232	35.6057	40.4312	49.9689	16.4192	15.7052
	8	1.1778	35.5847	40.8261	49.8879	16.4192	15.7104
	12	1.1599	35.4611	40.8344	49.7480	16.4216	15.7337
	16	1.1548	35.2866	41.0475	49.7116	16.4299	15.7746
	20	1.1433	35.1749	41.1145	49.6782	16.4781	15.8445
	24	1.1342	35.0972	41.1306	49.1616	16.5175	15.9688

Note: Results are presented as the mean \pm standard deviation (n=3).

Table 3: Correlation Coefficient (R^2) of Unsaturated Fatty Acids (18:3, 18:2 and 18:1) and Parameters of Oil Oxidation (PV, p -AV and TBARS) in Peanut Oil and Flaxseed Oil Samples

	Peanut Oil			Flaxseed Oil		
	Control	Rosemary (Powdery)	Rosemary (Oily)	Control	Rosemary (Powdery)	Rosemary (Oily)
18:3 vs. PV	-0.848	-0.927	-0.844	-0.930	-0.980	-0.971
18:2 vs. p -AV	-0.953	-0.995	-0.951	-0.757	0.790	0.837
18:2 vs. TBARS	-0.961	-0.993	-0.959	-0.801	0.919	0.863
18:1 vs. p -AV	-0.936	0.927	0.838	-0.512	0.805	0.818
18:1 vs. TBARS	-0.974	0.898	0.898	-0.493	0.938	0.880

percentage changes in linoleic acid (18:2) was maximum in control and minimum in stabilized oil with powdery rosemary extract in each interval, indicating its highest antioxidant efficiency. The results from changes in fatty acids composition was similar that in oxidative stability. The order of antioxidant efficiency was powdery rosemary extract > oily rosemary extract. Furthermore, an interesting phenomenon was observed in the changes of oleic acid (18:1), it was shown that the percentage of oleic acid decreased for the control in the 24 days of storage, whereas it increased for all the stabilized samples. The order of increase observed was the same as that of antioxidant efficiency, it was indicated that antioxidants inhibits the oxidative decomposition of oleic acid and samples stabilized with powdery rosemary extract had the highest antioxidant efficiency. As to the flaxseed oils, the same order of antioxidant efficiency was obtained from the results of changes in unsaturated fatty acids (18:3, 18:2 and 18:1). However, the change trends in relative fatty acid composition for the control and the stabilized samples were opposite both for linoleic acid (18:2) and oleic acid (18:1) as shown in Table 2.

DISCUSSION

The correlations coefficient (R^2) of relative contents of 18:3, 18:2, 18:1 in oils and the peroxide value (PV), p -anisidine value (p -AV) and thiobarbituric acid-reactive substances assay (TBARS) in the storage of 24 days are shown in Table 3. For the control of peanut oils, the correlations were all negative. The peroxide values did not correlate well with the 18:3 contents ($R^2=0.848$, $p<0.05$), whereas the other two indices of oxidative stabilities correlated well with both the 18:2 contents ($R^2>0.953$, $p<0.05$) and the 18:1 contents ($R^2>0.936$,

$p<0.05$), indicating the 18:2 content and 18:1 content can closely predict the oxidative stabilities. Compared to the control, positive correlations between the 18:1 contents of oils and p -AVs and TBARSs for all stabilized peanut oils after storage at 60°C for 24 days. It was indicated all the antioxidants inhibited the oxidative decomposition of 18:1, which delayed the oxidative rate of peanut oils. For the control of flaxseed oils, highly negative and significant correlations were found between the 18:3 contents and PVs ($R^2=0.930$, $p<0.05$). However, the 18:2 contents and 18:1 contents did not correlate well with p -AVs and TBARSs though they were also negative. Interestingly, when compared to the control of flaxseed oils, positive correlations were obtained between the 18:2 contents and 18:1 contents of flaxseed oils and p -AVs and TBARSs for all stabilized samples with antioxidants. It was assumed that antioxidants inhibited the decomposition of both 18:2 and 18:1 in flaxseed oils. Furthermore, the 18:3 contents and PVs were highly negative correlated with each other for all stabilized flaxseed oils.

CONCLUSION

As shown by the results of PV, AV, p -AV and TBARS, rosemary extract had significant effectiveness in anti-oxidation of peanut oil and flaxseed oil, and powdery rosemary extract was more effective than oily rosemary extract though with no significant differences. In the accelerated storage up to 24 days, the oxidation of flaxseed oil samples were significantly faster than that of peanut oil samples as flaxseed oil contained more 18:3 than did the peanut oil. Moreover, natural antioxidants delayed the oxidative rate of both peanut oils and flaxseed oils by inhibiting the decomposition of some of the unsaturated fatty acids. For controls of both peanut oil and flaxseed oil, negative correlations

(18:3 vs. PV; 18:2 vs. p-AV; 18:2 vs. TBARS; 18:1 vs. p-AV; 18:1 vs. TBARS) were found between each other. However, for stabilized samples, positive correlations (18:1 vs. p-AV; 18:1 vs. TBARS) were found in peanut oil, and positive correlations (18:2 vs. p-AV; 18:2 vs. TBARS; 18:1 vs. p-AV; 18:1 vs. TBARS) were observed in flaxseed oil. It was indicated that rosemary extracts effectively inhibited the oxidation of both oils mainly by retarding the decomposition of 18:1 in PO samples, while by retarding the decomposition of 18:2 and 18:1 in FO samples. Therefore, the differences in fatty acids composition of peanut oil and flaxseed oil resulted in the differences in their oxidative stability and different mechanisms of anti-oxidative effectiveness of antioxidants.

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