Effect of using rice bran oil in spreadable fats preparation on quality criteria during cold storage

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ABSTRACT

The current study aims to the possibility of using rice bran oil (RBO) instead of soybean oil (SO) in preparing spreadable fat and its effect on oxidative stability, sensory evaluation and chemical properties during the cold storage period. The results have shown that the use of RBO leads to an increase in the ability to persist against oxidation and increases with the increase in the amount of oil used in the samples with no artificial antioxidants added. Also, no significant differences were noticed between the mean sensory values that were evaluated between the prepared samples and the control sample as well as the commercial sample. The results also showed an increase in free fatty acids (at different rates) in all spreadable fat samples included in the study during the cold storage period. The fatty acids were relatively higher in the case of RBO, and the increase continued gradually throughout the experimental storage period. Also, there was a gradual increase in the peroxide value of the extracted fats during the cold storage period, and then a gradual decrease occurred after that in all treatments except for the commercial sample and the treatment 80% of total fat (used in preparing RBO) as there was no decrease until the end of the storage period.

Keywords: rice bran oil, spreadable fat, oxidative stability, peroxide value, thiobarbituric acid

INTRODUCTION

Rice (*Oryza sativa* L.) is an important cereal crop and a staple food for more than half of the world population (Wani *et al.,* 2012). Rice bran is a by-product of the rice milling industry. Bran is the part between paddy husk and endosperm representing 10% of the rough rice kernel and is obtained during polishing of rice. Rice bran is a brown layer presented between rice grain and the outer husk of paddy that is rich in protein, oil and other nutrients. It is one of the major by-products of rice production and it has huge potential to be exploited as a substrate for the production of value-added products using biotechnological tools such as fermentation as well as applications in foods, health and cosmetic industries (Amarasinghe *et al.,* 2009; Pourali *et al.,* 2010; Wang, *et al.,* 2015). The quality properties of many food products strongly depend upon the physical properties of fat and oils. Physical properties of fat and oils are of critical importance in determining their use (Devi and Khatkar, 2017). RBO could be used as an edible material as well as for industrial purposes and perceived good stability but the only problem is associated with the oil to reduce the free fatty acids (Anwar *et al.,* 2006).

RBO, extracted from the germ and the inner husk of the rice grain, is rich in gamma oryzanol (Patel and Naik, 2004 and Tuncel and Yilmaz, 2011). γ-oryzanol is one of the important components in RBO having nutraceutical properties and can be useful in potential pharmaceutical and cosmeceutical preparations. It's a mixture of ferulic acid ester of sterol and triterpene alcohols. It occurs in RBO at a level of (1:2%), and it also serves as a natural antioxidant. γ-oryzanol, a major component of RBO, was used to decrease plasma cholesterol, platelet aggregation, cholesterol absorption from cholesterol-enriched diet and aortic fatty streaks. Other than the promising cholesterol-lowering effects, γ-oryzanol may have other pharmacological effects such as regulation of the estrous cycle, growth accelerating action and the ability to promote skin capillary circulation (Sugano *et al.,* 1999).

RBO was added for food systems to increase of oxidative stability and the same times to improve the nutritional and health benefit values of food (Godber, 2009). It is unique among edible oils due to being a rich source of commercially and nutritionally important phytochemicals such as polyphenol, tocopherol, γ-oryzanol, lecithin, squalene, phytosterol, tocotrienol, among others. Many studies on human and animals have shown that RBO is effective as some other vegetable oils that are rich in polyunsaturated fatty acids for lowering plasma cholesterol level and lipoprotein profile. In some cases, RBO is beneficial to lower plasma cholesterol more effectively, and this effect can be attributed to the occurrence of these other specific components in RBO (Lai *et al.,* 2019). The current study aims to evaluate the possibility of using RBO instead of SO in preparing spreadable fat and to estimate its effect on oxidative stability, sensory evaluation and

chemical properties during the cold storage period.

MATERIALS AND METHODS

Materials

Rice bran oil (RBO)

The refined, bleached and deodorized RBO used in the present study was obtained in 2018 from Al-Bustan Company for Investment and Commercial Development, Izbat Al-Nakhl, Cairo, Egypt.

Palm and soybean oils

Refined, bleached and deodorized palm (PO) and soybean (SO) oils used in the present study were obtained in 2018 from Arma Food Industries, 10th of Ramadan City, Egypt.

Additives

Corn starch (thickening agent), obtained from Tag El Melouk Food Industries – 6th of October City, Egypt, sodium alginate (thickening agent) obtained from Morgan Chemical Industries Company, 10th of Ramadan City, Egypt. Whey protein powder (3% total protein, stabilizer) was obtained from DV Nutrition – Hoogeveen – Holland, skim milk powder (emulsifying agent) obtained from Arabe Cultivators Company, Giza, Egypt, soybean lecithin (emulsifying agent) imported from Emulgrain S.A., Argentina, glycerol monostearate (monoglyceride, emulsifying agent) obtained from Oleon a Natural Chemistry, Belgium, glycerol monostearate (antioxidant) obtained from AWA for Food Additives, Alexandria, Egypt, potassium sorbate (preservative) obtained from El-Gomhouria Trading Chemicals and Drugs Company, Cairo, Egypt. The butter flavor was purchased from the Egyptian French Company for Investment, industrial area, Ismailia, Egypt and was used in the spreadable fat formulation.

Methods

Preparation of spreadable fat

Spreadable fat contains 40, 60 and 80% total fat were formulated according to the method described by Cheng *et al.* (2008), Goli *et al.* (2009), and Lumor *et al.* (2010), with some modifications and their ingredients were shown in Table (1). Fat phase comprised oils and additives soluble in fat (soy lecithin, glycerol monostearate and ascorbyl palmitate) were mixed in a stainless steel vessel, gently warmed at 40-50°C with continuous steering until complete solubilization and homogenecity were achieved (5-10 min). Using another stainless-steel vessel, aqueous phase (boiling water, corn starch, skim milk powder, sodium alginate, potassium sorbate and whey protein powder were mixed using an electric mixer at ambient room temperature until completed to solubilization and homogenecity (1-2 min). Blessing vessel fat phase in an ice bath to accelerate crystallization, soluble aqueous phase it was added slowly into the fat phase and blended with a home mixer (mixing at low speed for 10 minutes followed by high speed for 5 minutes) until complete homogenization occurred and the suitable emulsion was achieved where the prepared spread had semi-solid texture. Polypropylene containers (50g capacity) were completely filled with the prepared spreadable fats, stored at refrigeration temperature (5-7°C). Storage experimental periods for 24 weeks were carried out at (5±1°C) where samples were analyzed periodically every two weeks during the storage period.

Analytical methods

Physical properties

Refractive Index, relative viscosity value, melting point and texture were determined according to the method described by A.O.A.C. (2011).

Chemical properties

(a) Free fatty acids content (% FFA)

The **%FFA** (as oleic acid %) of tested treatment samples were determined according to the method of IUPAC (1987). A mixture of absolute ethanol and diethyl ether (1:1 v/v) was carefully neutralized with 0.10m NaOH solution using 1% phenolphthalein indicator. 5 g of the treatment samples were dissolved in 50 ml neutralized ethanol diethyl ether solvent and titrated with 0.10 M KOH with constant shaking until a pink color persist for 15 sec. **%FFA** were calculated as of % oleic acid.

(b) Iodine value (IV)

The Iodine value was calculated from the fatty acids composition of tested oils according to Ham *et al.* (1998).

(c) Determination of gamma oryzanol content

Gamma oryzanol content of the RBO sample was determined by spectrophotometric method according to the method described in Codex (2015) with spectrophotometer absorption measurements at the wavelength 314 nm in 1-cm quartz cuvette. reference cuvette with n-Heptane.

Determination of fatty acids composition

The fatty acids (FAs) of tested oil samples were determined as methyl ester by gas-liquid chromatography (HP 6890, London. UK.) operated under the following conditions: The methyl ester samples were using MeOH (20%) as a methylating, Detector, flame ionization; column, capillary, 30.0 m X 530 μm, 1.0 μm thickness, polyethylene glycol phase (INNO Wax); N2 with flow rate, 15 ml/min. with average velocity 89 cm/s (8.2 psi), H_2 flow rate, 30 ml/min.; Air flow rate, 300 ml/min.; Split ratio, 8:1, Split flow, 120 ml/min.; Gas saver, 20 ml/min.; Detector temp. 280 ^{0}C; Column temp. 240 ^{0}C; Injection temp. 280 ^{0}C.; Programmed temp. starting from 100 ^{0}C to reach a maximum of 240 ^{0}C (with 10 ^{0}C/min rising; then held at 240 ^{0}C for 10 min.) was used for eluting the fatty acid methyl esters.

Determination of Trans- fatty acids:

The trans-fatty acid content in oil blends was determined by using spectrophotometer (UV-2100, Shimadzu. Corporration, Tokyo, Japan) according to the method of Martinez *et al.* (2010).

The oxidative stability

(a) ***Rancimat test***

The oxidative stability of spreadable fat samples was determined by using automated Rancimat (Metrohm Ud. CH 9100 Herisau, Switzeland, model 679) according to Tsaknis *et al*. (1999).

(b) Peroxide value (PV)

The peroxide value of investigated samples was determined according to the method described in A.O.A.C. (2011).

(c) Thiobarbituric acid value (TBA)

The TBA of tested samples was determined according to the method described in Pearson (1976).

Organoleptic Evaluation

Seven sensory characteristics of studied spreadable fat samples after preparation were evaluated. These include color, odor, taste, spreadability, texture, appearance and overall acceptability. Ten members from the food science and technology department, Faculty of Agriculture, Al-Azhar University comprise panelist group. A scoring list of 10 points for each sensory characteristic was used according to Kolanowski and WeiXβrodt (2007).

Storage experimental periods

The commercial sample and four prepared spreadable fats were subjected to experimental storage periods at refrigeration temperature (5±1°C) for 24 weeks. During storage, every two weeks, spreadable fat samples of each treatment were withdrawn and subjected to chemical tests.

Statistical analysis

One-way analysis of variance (ANOVA) using SPSS 20 was performed on all experimental data sets, Post-hoc multiple comparisons were carried out by Duncan's Multiple Range Tests were done according to Helwing (1983).

RESULTS AND DISCUSSION

Physicochemical properties of commercial and prepared spreadable fat samples:

The physicochemical properties of commercial and prepared spreadable fat samples were presented in Table (2). The RI was related to the ease with which light passes through the fat. Temperature and degree of saturation affect the value, RI of spreadable fat was 1.4610, 1.4612, 1.4645, 1.4647 and 1.4650 of commercial, control, 40%, 60% and 80% total fat samples, respectively, the RI of oils depends on their molecular weight, fatty acid chain length, degree of unsaturation and degree of conjugation.

Results in Table (2) indicated that as the percent of total fat increased in spreadable fat, a parallel increase in melting point was noticed. The melting point is an important physical characteristic of fatty compounds which is useful for identification and important in many technological applications of fatty materials. The melting points of fatty acid increase with increasing chain length and decrease as the acids become more unsaturated, and melting points were found to be 35.5 and 32.0 ºC for commercial and control samples, respectively; 31.5, 33.0 and 33.5 ºC for treatments contained 40, 60 and 80 % total fats, respectively. Variation of melting point might be due to variation in total fat% (Hui, 1996; Shin *et al*., 2010).

From Table (2), it's clear that the four formulated samples under study exerted low values of % FFA ranging between (0.119 and 0.692% as oleic acid) for commercial sample and 40% total fat, respectively, while PV (0.476 – 1.788 meqO_2/kg oil) for control and commercial samples respectively, regarding TBA value (0.277-0.520 mg malonaldehyde/Kg oil) for 40 and 80% total fats respectively. These figures

agreed with those adopted by Egyptian Standard Specifications (2007) (No. 6374) for spreadable fat and blended spreads that indicate the good quality of formulated spreadable fat.

Results in Table (2) reveals that as total fat% of formulated spreadable fat was increased, viscosity value was also elevated. Therefore, commercial, control and 80% total fat samples showed the highest viscosity values (0.620, 0.600 and 0.610 P., respectively), while treatment contained 40 and 60% total fat exerted lower values (0.499-0.568 P., respectively). These findings concede with viscosity values of oils used in the formulation as % of PO (had very high viscosity values compared to the other two oils) was evaluated as total fat content of spreadable fat was increased. A similar trend was found by Jenab and Temelli (2011). The texture of spreadable fat is one of the major properties of these products (Krawczyk *et al.*, 1996; Smewing, 1999).

Bourne (2002) cited that texture properties of foods are physical properties that are sensed by the feeling of touch and were measured objectively as a function of stress, time and distance. However, sensory evaluation is time-consuming, expensive and often of poor reproducibility. The texture is a multi-parameter attribute that is measurable by sensory and instrumental methods (Adhikari *et al.*, 2001; Kolanowski *et al*., 2006).

The textural properties (firmness or hardness) of formulated spreadable fat samples were objectively determined. Results in Table (2) showed that a total fat % of spread increases, hardness values was also markedly increased. So, spread sample of 60% and 80% total fat had the highest values (13.7 and 14.4 N for hardness, respectively) compared with commercial and control samples. These high value for texture parameter makes such spreadable fat to be more difficult to spread compared to other treatment of lower total fat content. On the other hand, the spread sample of the lowest fat content (40%) exerted the lowest firmness value (8.9 N) because they were formulated with less fat. Bourne (2002) mentioned that texture properties of foods are physical properties that are sensed by the feeling of touch and are measured objectively as a function of stress, time and distance. However, the sensory evaluation is time-consuming, expensive and often of poor reproducibility. It is now generally recognized that texture is a multi-parameter attribute measurable by both sensory and/or instrumental methods (Adhikari *et al.*, 2001 and Kolanowski *et al*., 2006).

Fatty acid profiles of commercial and prepared spreadable fat samples

An ideal edible oil should contain SFA, MUFA and PUFAs in proportions of 1:1.5:1 ratio to meet the recommended intake of fatty acids. However, this is not the case in practical terms as all the edible oils differ in their fatty acid composition. The RBO has almost similar ratio of fatty acids as recommended by the WHO for lowering blood cholesterol levels (Lai *et al.,* 2012; Friedman, 2013). Fatty acid composition of commercial and prepared spreadable fat samples was tabulated in Table (3), when fatty acid profile of lipid extracted from spreadable fats were carried out, 14 fatty acids (9 SFAs and 5 USFAs) were detected in commercial sample, while the fatty acid profile of control and prepared spreadable fat samples was carried out, 12 fatty acids (6 SFAs and 6 USFAs) were detected in all other treatment. Commercial sample had a relatively high content of TSFAs (50.63%) (Table 3).

The fatty acid C16:0 showed the highest value among SFAs as its content reached 43.75% of total fatty acids, while all other treatment within (24.42-27.53%) in treatment content RBO. The USFAs (49.37%), C18:1 and C18:2 represent the predominant UFAs in commercial sample (38.45 and 10.15%, respectively), while other treatments content RBO as their contents reached to within (69.61–72.24%), C18:1 and C18:2 represent the predominant USFAs showed the highest value (40.97-28.15%, respectively). It is combined with other active ingredients in the oil due to the cholesterol-lowering effect, the PUFAS in RBO exert greater hypolipidemic activities as compared to other vegetable oils containing linoleic acid and therefore may help to lower the cardiovascular risk (Friedman, 2013). Studies, however, suggest that cholesterol-lowering properties of RBO could mainly be due to unsaponifiable fraction of bioactive components rather than because of fatty acid composition (Abumweis *et al.,* 2008; Liang *et al.,* 2014).

It is evident from Table (3) that the commercial sample and four samples that were installed in this study are free of trans fatty acids.

Rancimat test

The spreadable fats stability index (induction period) was defined as the point of maximum change of the rate of oxidation (Wagner *et al.*, 2000). Both peroxide values and

stability tested values by rancimat were used to given indication about the early stage of oil oxidation where hydro peroxides were formed (Zhang *et al.*, 2010). Lipid oxidation is a degradation process considered to be a major cause of quality deterioration of spreadable fat products. It imparts rancid and unpleasant flavors to the products and thus decreases their organoleptic value. Hence, oxidative stability of shortenings, margarine and oils was a concern to bakers and snack food fryers. Therefore, the oxidative stability of lipid extracted from tested spreadable fats was determined immediately after formulation.

Results in Table (4) showed that spreadable fat samples containing RBO showed high oxidative stability than those blends for free RBO may be due to containing RBO of nutrient composition, these results were nearly in agreement with (Chotimarkorn and Silalai 2008; Paul *et al.* 2012). As presented data from Table (4), the highest value of the commercial sample (59.02 h), while the lowest value (22.78 h) was recorded for control sample. On the other hand, the values (27.22, 31.68 and 37.06 h) were recorded for 40%, 60% and 80% total fats, respectively.

It could be concluded that differences of oxidative stability between all tested spreadable fat samples may be due to the natural antioxidants content in spreadable fat samples (especially, the gamma oryzanol occurs RBO as a major component at the level of (1:2%) and it serves as a natural antioxidant and the degree of unsaturation and number of double bonds in triglycerides for oils used in the processing of these spreadable fats (Anwar *et al*., 2003).

In general, the use RBO in preparing spreadable fats leads to improvement in the stability against oxidation, which leads to a prolongation of the storage period without the use of industrial antioxidants.

Sensory evaluation of spreadable fat samples

The sensory parameters such as color, flavor, taste, texture, and overall acceptability of any food product depends on the extent of oxidation of fats and oils in food due to the formation of aldehydes, ketones and peroxides, (Gupta, 2005). A sensory evaluation was carried out by the senses of taste, smell, touch, and hearing when food was eaten. The complex sensation that results from the interaction of senses was used to measured food quality in programs for quality control and new product development. The sensory evaluation results in Table (5) showed that all treatments were accepted by the panelists.

The organoleptic quality criteria of edible fat and oil products was correlated significantly with their chemical constituents responsible for color and flavor characteristics and with the chemical reactions and the deterioration that take place in them during their processing, storage and utilizing for food cooking. In addition, the sensory evaluation results for edible fat and oil products providing with information about palatability and consumer acceptability of these lipids and foodstuffs cooked or processed as well as with an expectation for their possible utilization in edible or industrial purposes. Also, sensory evaluation was an important tool that links product attributes with consumer preferences. The organoleptic characteristics, including color, taste, odor, spreadability, texture, appearance and overall acceptability of formulated spreadable fat samples under study were evaluated. The obtained results were statistically analyzed and recorded in Table (5).

The scheduled results showed no significant differences ($p \leq 0.05$) for the values of the seven characteristics evaluated for the treatments compared with the commercial and control samples. Therefore, the mean values recorded for 40%, 60% and 80% total fat samples were (7.69, 7.38, 7.53, 7.92, 7.46, 7.38 and 7.23); (8.00, 7.76, 7.92, 7.76, 7.69, 8.00 and 7.84); (7.69, 7.38, 7.69, 7.53, 7.57, 7.92 and 7.46) for the color, taste, odor, spreadability, texture, appearance, and overall acceptance, respectively. On the other hand, showed the commercial sample as well as a control sample (containing 80% total fat) corresponding mean values of (8.00, 7.46, 7.95, 7.92, 7.69, 8.00 and 7.46) and (7.94, 6.46, 7.15, 7.53, 7.19, 7.76 and 7.15), respectively.

The use of RBO in the formulation process showed no significant differences ($p \leq 0.05$) in the mean value of the sensory properties evaluated. These results were nearly in agreement with (Shaik *et al*., 2017).

Changes in chemical properties of spreadable fat samples

The effect of the cold storage period (5±1ºC) on some chemical attributes formulated spreadable fat samples was studied. The cold storage experimental period was extended for 24 weeks (the shelf life of spreadable fat samples as recommended by the Egyptian Standard Specifications (2008) (No. 2613-2). During storage, every two weeks, a sample representing each treatment was withdrawn

and tested for its free fatty acid%, peroxide value and TBA (as chemical properties).

Free fatty acids (FFA%)

Hydrolytic processes lead to the formation of FFA (%) by splitting of acylglycerols that can affect flavor. FFA% is an important quality indicator during the processing and storage of fat and oils. Generally, fat and oils were susceptible to enzymatic hydrolysis; the FFA (%) formed varies with age and storage. FFA (%) determination is a quality marker for establishing the extent of hydrolytic rancidity in fat and oils. Normally, fatty acids (FAs) are found in triglyceride form, however, during processing, the FAs may get hydrolyzed into FFA (Choudhary and Grover, 2013). The commercial sample and four formulated spreadable fat samples initially and during storage were subjected for FFA (%) analysis and the values obtained were listed in Table (6). As shown in Table (6), cold storage increases FFA% of all studied treatments. Increase (at different levels) progressively occurred as the storage period prolonged. At the end of the storage period (24 weeks) high significant ($p \leq 0.05$) increases were noted for all studied samples. For example, 80% total fats treatment had FFA% content of 0.281% at zero time that increased to 1.122% at the end of the storage period, while commercial control samples (without RBO) showed corresponding values of 0.119, 0.131% and 0.906, 0.614% at the same periods, respectively.

From the same Table (6), it could be observed that treatments containing RBO showed high FFA% compared with their control treatment. This finding was noted at the beginning and throughout the whole storage period. For example, FFA% of (40% total fat) showed FFA% content of 0.692% at zero time that increased to 0.920% after 24 weeks of cold storage, as well (60% total fat) showed FFA % content of 0.518% at zero time that increased to 0.860% after 24 weeks of cold storage at the same periods of storage, respectively. This result may be related to the higher FFA% content of RBO compared to PO and SO. These findings were found in general agreement with Patange *et al.* (2013), who found increasing of FFA% and TBA value of spreadable fats during the cold storage period. The increase in FFA% could be attributed to the hydrolysis of triglycerides by the moisture from the oils and due to oxidation. The presence of moisture can be a result of other deteriorative chemical reactions leading to hydrolytic rancidity. Przybylski and Daun (2001) ascribed the rise in FFA% content during cold storage mainly to hydrolysis of triglycerides and the cleavage of secondary oxidation products formed during cold storage.

General, FFA% of all studied samples during the whole storage periods were found within the range published by Egyptian Standard Specifications (2007) (No. 6374) for spreadable fats and blended spreads.

Peroxide value (PV)

PV is known as an indication of the extent of forming the hydroperoxides, the primary products of lipid oxidation in foodstuffs, which are generally referred to as peroxides. The PV has often been seen to reach a maximum value and can decrease as lipid oxidation proceeds and the hydroperoxides breakdown at a faster rate than formation to the secondary oxidation products (Cecilia *et al*., 2005). PV was a measure of oxidation during storage and the freshness of the lipid matrix (Malheiro *et al.*, 2013). In addition, it gives important information about lipid autoxidation. PV measures only the earlier stage of oxidation and primary oxidation products (Atinafu and Bedemo, 2011; Srivastava and Singh, 2015). In general, the lower the PV, the better the quality of the oil. However, PV decreases as secondary oxidation products appear (Chakrabarty, 2003). Changes in PV (meqO_2/kg oil) of commercial and prepared spreadable fat samples was determined during cold storage (5±1°C) for 24 weeks and listed in Table (7).

The PV of all tested spreadable fat samples was progressively increased during cold storage in all transactions during the cold experimental storage period, then it decreased again until the end of the experimental storage period, except for the commercial sample and the 80% total fat treatment, which did not decrease until the end of the storage period, as that value at the beginning of the storage period was 1.788 and 0.865 meqO_2/kg oil and increased that value until it reached 11,549 and 11,808 meqO_2/kg oil, respectively, at the end of storage period (Table 7). This may be due to the use of industrial antioxidants in the commercial sample and the containment of RBO on natural antioxidants, such as (gamma oryzanol, tocopherol and tocotrienol) in which RBO was used in a rate greater than 40 and 60 % total fat treatments, while we find that the control 2 sample in which SO was used, the PV increased until week 14 and then a sudden decrease at the end of experimental storage period, this may be due to the SO contained a high of polyunsaturated fatty acids and when replacing SO with RBO, we found that the PV reached it

was highest value at week 20, and then a decrease occurred after that. During storage, autoxidation takes place and PV reaches a maximum followed by a decrease at more advanced stages varying according to the fatty acids composition and conditions of oxidation (Frankl, 2005). Also, Alexa et al. (2010) reported that oxidation of the fat phase was evident by significant increases in PV of all spreads on storage.

General, comparison with values found in Egyptian Standard Specifications (2007) (No. 6374) for edible spreadable fats and blended spreadable fats, it can be said that the studied differences recorded values of PV within the limits recommended for human consumption of 10 meqO_2/kg oil at different periods during storage.

Thiobarbituric acid value (TBA):

TBA value was condensed as an indication for the amount of malonaldehyde which has the most predominant secondary oxidation products of food lipids. It has a good chemical constant for quality assurance and measuring the extent of secondary oxidation of edible lipids (Rodriguez-Estrada *et al*., 1997). During lipid oxidation, hydro peroxides, the primary reaction products, decompose to produce secondary oxidation products aliphatic aldehydes (saturated and unsaturated), alcohols, ketones, acids and hydrocarbons which were more stable during the heating process, responsible for off-flavors and off-odors of edible oils. TBA value of commercial, control sample and the treatments were displayed in Table (8), showed a significant increase in TBA values could be noticed between the initial and final storage periods in the treatments indicated the development of flavor but there were not large enough to cause perceptible changes up to 10 weeks of cold storage periods at (5±1°C).

From Table (8) the value ranged from 0.332 to 1.888 mg malonaldehyde/kg oil in commercial sample for a period of 24 weeks, which recorded the lowest values. Control sample recorded a value of 0.387 initially and 3.253 mg malonaldehyde/kg oil after 24 weeks of storage, which recorded the highest values between treatments, this may be due to the fact that it contains SO unlike other treatments. Concerning 40% total fat recorded a value of 0.277 initially and 2.765 mg malonaldehyde/kg oil after 24 weeks of storage, while 60% and 80% total fat TBA value ranged from 0.407 to 2.407 mg malonaldehyde/kg oil and 0.520 to 2.027 mg malonaldehyde/kg oil respectively, for a period of 24 weeks. These findings were found in general approximately in agreement with the results of Patange *et al.* (2013) and Lund *et al.* (2008) who reported that the amount of malonaldehyde released (as secondary oxidation product) increases during storage time.

General, tested spreads had reasonable TBA values up to 24 weeks of cold storage as Greene and Cumuze (1982) suggested that the border line of TBA value for the edible fats and oil for human consumption is 10.0 mg malonaldehyde per kg of lipid, after which the lipids might be completely oxidized and should not be used for human consumption.

CONCLUSION

It can be concluded from the results obtained that the use of RBO in preparing spreadable fats leads to improvement in the stability against oxidation, which leads to a prolongation of the storage period without the use of industrial antioxidants. Also, the use of RBO in the formulation process was recorded no significant differences ($p \leq 0.05$) in the mean value of the sensory properties evaluated. FFA% and PV of all tested spreadable fats samples during the whole storage periods were found within the limits recommended for human consumption published by Egyptian Standard Specifications (2007) (No. 6374). The tested spreadable fats samples had reasonable TBA values up to 24 weeks of cold storage, after 24 weeks which the lipids might be completely oxidized and should not be used for human consumption.

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Table 1. Ingredients of formulated spreadable fat samples.

Ingredient %	Spreadable fat			
	Control sample without RBO	With RBO		
		40% total fat	60% total fat	80% total fat
Fat phase				
Palm oil	40.00	7.70	11.80	15.90
Soybean oil	39.50	-	-	-
Rice bran oil	-	30.80	47.20	63.60
Glycerol monostearate	0.60	1.00	0.60	0.60
Soybean lecithin	0.20	0.50	0.40	0.20
Ascorbyl palmitate	0.02	0.02	0.02	0.02
Butter flavor	0.03	0.03	0.03	0.03
Aqueous phase				
Water	16.00	40.00	20.00	16.00
Potassium sorbate	0.15	0.20	0.15	0.15
Skim milk powder	1.00	4.40	5.00	1.00
Sodium alginate	1.00	1.00	1.00	1.00
Whey protein	1.00	4.50	4.00	1.00
Corn starch	1.00	9.85	9.30	1.00

RBO: Rice bran oil.

Table 2. Physicochemical properties of commercial and prepared spreadable fat samples.

Quality Parameter	Commercial sample	Spreadable fat			
		Control sample without RBO	With RBO 40% total fat	With RBO 60% total fat	With RBO 80% total fat
Refractive index	1.4610	1.4610	1.4645	1.4647	1.4650
Melting point (ºC)	35.5	32.0	31.5	33.0	33.5
Viscosity (P)	0.620	0.610	0.499	0.568	0.600
FFA (as oleic acid %)	0.119	0.131	0.692	0.518	0.281
PV (meq. O_2/kg oil)	1.788	0.476	0.971	0.479	1.325
TBA value (mg malonaldehyde/Kg oil)	0.332	0.387	0.277	0.407	0.520
IV (I_2/100 g)	51.33	89.29	83.03	85.44	88.50
Texture Firmness (N)	16.3	15.0	8.9	13.7	14.4
Gamma oryzanol (mg/100gm oil)	0	0	154	236	318

RBO: Rice bran oil, FFA: free fatty acid, PV: peroxide value,TBA: thiobarbeturic acid, IV: iodine value.

Table 3. Relative percentage of fatty acid of commercial sample and prepared spreadable fat.

Fatty acids	Commercial sample	Spreadable fat			
		Control sample without RBO	With RBO 40% total fat	With RBO 60% total fat	With RBO 80% total fat
$C_{6:0}$	0.06	ND	ND	ND	ND
$C_{8:0}$	0.07	ND	ND	ND	ND
$C_{10:0}$	0.34	ND	0.18	0.06	ND
$C_{12:0}$	1.12	0.12	0.31	0.14	0.05
$C_{14:0}$	0.16	0.61	1.08	0.72	0.51
$C_{16:0}$	43.75	27.53	26.28	25.39	24.42
$C_{16:1}$	0.19	0.13	0.25	0.21	0.19
$C_{17:0}$	0.12	0.09	ND	ND	ND
$C_{18:0}$	4.85	4.03	3.46	3.07	2.63
$C_{18:1}$	38.45	32.83	39.30	39.86	40.97
$C_{18:2}$	10.15	31.26	26.97	28.07	28.15
$C_{18:3}$	0.20	2.60	0.88	1.08	1.66
$C_{20:0}$	ND	0.35	0.80	0.78	0.76
$C_{20:1}$	0.38	0.19	0.36	0.40	0.43
$C_{22:0}$	0.16	0.26	0.22	0.23	0.23
TSFAs	50.63	32.99	32.33	30.39	28.60
MUFAs	39.02	33.15	39.91	40.47	41.59
PUFAs	10.35	33.86	27.85	29.14	29.81
TFAs	0.00	0.00	0.00	0.00	0.00

TSFAs: total saturated fatty acids, PUFAs: polyunsaturated fatty acids, TFAs: trans fatty acids, MUSFAs: monounsaturated fatty acids, RBO: rice bran oil, ND: not detected.

Table 4. Oidative stability of spreadable fat samples.

Oxidative stability at 100 ˚C		Induction period/h	Induction period in months (Validity period)
Spreadable fat	Commercial sample	59.02	20.98
	Control sample without RBO	22.78	8.10
	With RBO: 40% total fat	27.22	9.67
	With RBO: 60% total fat	31.68	11.26
	With RBO: 80% total fat	37.06	13.17

RBO: Rice bran oil.

Table 5. Sensory evaluation of spreadable fat samples.

Quality Parameter	Spreadable fat (M±SD)				
	Commercial sample	Control sample without RBO	With RBO 40% total fat	With RBO 60% total fat	With RBO 80% total fat
Color	8.00^{a}±1.29	7.94^{a}±1.39	7.69^{a}±0.94	8.00^{a}±1.29	7.69^{a}±1.25
Taste	7.46^{a}±0.59	6.46^{a}±1.98	7.38^{a}±1.98	7.76^{a}±0.59	7.38^{a}±1.44
Odor	7.95^{a}±1.40	7.15^{a}±1.40	7.53^{a}±0.87	7.92^{a}±1.25	7.69^{a}±1.54
Spreadability	7.92^{a}±0.95	7.53^{a}±1.08	7.92^{a}±0.95	7.76^{a}±1.53	7.53^{a}±1.50
Texture	7.46^{a}±1.50	7.19^{a}±1.25	7.46^{a}±1.50	7.69^{a}±1.79	7.57^{a}±1.41
Appearance	8.00^{a}±1.52	7.76^{a}±1.36	7.38^{a}±1.04	8.00^{a}±1.52	7.92^{a}±1.25
Overall acceptability	7.46^{a}±1.26	7.15^{a}±1.28	7.23^{a}±1.16	7.84^{a}±0.89	7.46^{a}±1.26

(M±S.D) = Mean ± SD. Deviation, RBO: rice bran oil.

Table 6. Changes in free fatty acid (%) of spreadable fat samples during cold storage (5±1°C) for 24 weeks.

Storage period (weeks)	Free fatty acid % of spreadable fat				
	Commercial sample	Control sample without RBO	With RBO 40% total fat	With RBO 60% total fat	With RBO 80% total fat
0	0.119±0.00^{d}	0.131±0.02^{d}	0.692±0.15^{a}	0.518±0.05^{b}	0.281±0.02^{c}
2	0.127±0.00^{c}	0.156±0.06^{bc}	0.701±0.15^{a}	0.572±0.09^{a}	0.306±0.04^{b}
4	0.170±0.06^{c}	0.159±0.05^{c}	0.715±0.15^{a}	0.595±0.07^{a}	0.395±0.08^{b}
6	0.181±0.04^{c}	0.215±0.02^{c}	0.721±0.15^{a}	0.630±0.04^{ab}	0.490±0.15^{b}
8	0.189±0.05^{b}	0.249±0.02^{b}	0.757±0.11^{a}	0.645±0.04^{a}	0.591±0.15^{a}
10	0.277±0.06^{b}	0.264±0.02^{b}	0.770±0.10^{a}	0.655±0.04^{a}	0.634±0.16^{a}
12	0.366±0.06^{b}	0.308±0.05^{b}	0.779±0.10^{a}	0.681±0.05^{a}	0.712±0.24^{a}
14	0.420±0.07^{b}	0.401±0.04^{b}	0.789±0.10^{a}	0.695±0.05^{a}	0.752±0.23^{a}
16	0.459±0.05^{b}	0.455±0.05^{b}	0.824±0.09^{a}	0.711±0.06^{a}	0.772±0.24^{a}
18	0.517±0.01^{bc}	0.501±0.07^{c}	0.850±0.07^{a}	0.731±0.07^{ab}	0.807±0.23^{a}
20	0.658±0.05^{ab}	0.526±0.05^{b}	0.854±0.07^{a}	0.784±0.09^{ab}	0.845±0.22^{a}
22	0.798±0.08^{a}	0.560±0.06^{b}	0.878±0.10^{a}	0.799±0.01^{a}	0.913±0.21^{a}
24	0.906±0.07^{ab}	0.614±0.10^{c}	0.920±0.16^{ab}	0.860±0.06^{b}	1.122±0.14^{a}

(M±S.D) = Mean ± SD. Deviation. Values with different small letters in the same row are significantly different ($p \leq 0.05$). RBO: rice bran oil

Table 7. Changes in peroxide value (meq O_2/kg oil) of spreadable fat samples during cold storage (5±1˚C) for 24 weeks.

Storage period (weeks)	Peroxide value of spreadable fat				
	Commercial sample	Control sample without RBO	With RBO		
			40% total fat	60% total fat	80% total fat
0	1.788±0.10^{a}	0.476±0.10^{c}	0.571±0.01^{c}	0.749±0.00^{b}	0.865±0.00^{b}
2	2.233±1.00^{a}	1.288±0.99^{ab}	0.760±0.01^{b}	0.880±0.09^{b}	0.894±0.01^{b}
4	3.262±1.05^{a}	2.166±0.00^{b}	1.672±0.07^{bc}	1.524±0.02^{bc}	0.942±0.00^{c}
6	4.170±0.01^{a}	4.150±0.15^{a}	3.290±0.03^{b}	2.282±0.00^{c}	1.074±0.02^{d}
8	4.617±1.00^{b}	6.790±0.10^{a}	4.155±0.15^{b}	3.993±0.00^{b}	1.995±0.00^{c}
10	5.260±0.01^{b}	8.636±0.60^{a}	5.718±0.01^{b}	4.598±0.01^{c}	3.865±0.01^{d}
12	5.828±1.02^{b}	10.206±1.20^{a}	7.039±0.01^{b}	6.032±0.03^{b}	5.752±0.00^{b}
14	6.457±0.10^{d}	11.786±0.01^{a}	9.397±0.01^{b}	8.458±0.05^{c}	6.282±0.01^{e}
16	6.492±1.04^{d}	9.683±0.00^{ab}	10.429±0.03^{a}	9.256±0.05^{b}	7.657±0.00^{c}
18	8.450±0.05^{d}	8.994±0.00^{c}	11.699±0.26^{a}	10.771±0.00^{b}	8.428±0.00^{d}
20	9.642±0.12^{b}	7.691±0.00^{c}	10.295±0.01^{b}	11.967±1.00^{a}	9.656±0.01^{b}
22	10.331±1.00^{a}	6.302±0.30^{b}	9.992±0.00^{a}	10.656±0.00^{a}	10.896±0.09^{a}
24	11.549±0.01^{a}	5.967±0.05^{d}	8.466±0.90^{c}	9.635±0.03^{b}	11.808±0.00^{a}

(M±S.D) = Mean ± SD. Deviation. Values with different small letters in the same row are significantly different ($p \leq 0.05$). RBO: rice bran oil

Table 8. Changes in thiobarbituric acid value (TBA) (mg malonaldehyde/kg oil) of spreadable fat samples during cold storage (5±1˚C) for 24 weeks.

Storage period (weeks)	(TBA) value of spreadable fat				
	Commercial sample	Control sample without RBO	With RBO		
			40% total fat	60% total fat	80% total fat
0	0.332±0.003^{d}	0.387±0.003^{c}	0.277±0.001^{e}	0.407±0.003^{b}	0.520±0.003^{a}
2	0.408±0.001^{d}	0.435±0.003^{c}	0.372±0.002^{e}	0.455±0.001^{b}	0.532±0.002^{a}
4	0.487±0.002^{e}	0.549±0.001^{d}	0.585±0.004^{c}	0.603±0.001^{b}	0.643±0.001^{a}
6	0.535±0.005^{d}	0.708±0.001^{b}	0.668±0.001^{c}	0.766±0.004^{a}	0.761±0.002^{a}
8	0.659±0.001^{e}	0.894±0.004^{a}	0.878±0.003^{b}	0.848±0.003^{c}	0.837±0.004^{d}
10	0.717±0.001^{e}	1.186±0.000^{a}	1.054±0.002^{b}	0.972±0.008^{c}	0.955±0.004^{d}
12	0.842±0.003^{e}	1.312±0.003^{a}	1.283±0.003^{b}	1.252±0.004^{c}	1.026±0.002^{d}
14	0.988±0.000^{e}	1.816±0.004^{a}	1.675±0.005^{b}	1.596±0.003^{c}	1.217±0.002^{d}
16	1.018±0.002^{e}	1.936±0.006^{a}	1.855±0.004^{b}	1.763±0.003^{c}	1.387±0.002^{d}
18	1.252±0.002^{e}	2.489±0.001^{a}	2.237±0.001^{b}	1.916±0.005^{c}	1.502±0.003^{d}
20	1.484±0.010^{e}	2.757±0.007^{a}	2.465±0.005^{b}	2.165±0.004^{c}	1.684±0.004^{d}
22	1.577±0.007^{e}	2.938±0.007^{a}	2.603±0.003^{b}	2.272±0.002^{c}	1.912±0.002^{d}
24	1.888±0.002^{e}	3.253±0.116^{a}	2.765±0.003^{b}	2.407±0.006^{c}	2.027±0.001^{d}

(M±SD) = Mean ± Standard deviation. Values with different small letters in the same row are significantly different ($p \leq 0.05$). RBO: rice bran oil.

تأثير استخدام زيت نخالة الأرز في تحضير الدهون القابلة للفرد على دلائل الجودة خلال التخزين على البارد

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الملخص العربي

تهدف الدراسة الحالية إلى إمكانية استخدام زيت نخالة الأرز بدلاً من زيت فول الصويا في تحضير الدهون القابلة للفرد وتأثير ذلك على الثباتية ضد التأكسد، وصفات الجودة الحسية والخصائص الكيميائية خلال فترة التخزين. وقد أظهرت النتائج أن استخدام زيت نخالة الأرز يؤدي إلى زيادة القدرة على الثباتية ضد التأكسد بزيادة كمية الزيت المستخدم في إعداد العينات بدون إضافة مضادات الأكسدة الصناعية. كما أنه لا توجد فروق معنوية بين متوسط القيم الحسية التي تم تقييمها وذلك بين العينات التى تم اعدادها والعينة التجارية. كما أظهرت النتائج زيادة في الأحماض الدهنية الحرة (بمعدلات مختلفة) في جميع عينات الدهون القابلة للفرد خلال فترة التخزين على البارد، وقد كانت أعلى نسبيًا في حالة زيت نخالة الأرز واستمرت الزيادة تدريجياً طوال فترة التخزين. وأيضا كانت هناك زيادة تدريجية في قيم رقم البيروكسيد للدهون المستخلصة أثناء فترة التخزين على البارد، ثم حدث انخفاض تدريجي بعد ذلك فى جميع المعاملات باستثناء العينة التجارية والمعاملة 80% دهون كلية (المستخدم في اعدادها زيت نخالة الأرز) حيث لم يحدث انخفاض حتى نهاية فترة التخزين.

الكلمات الاسترشادية: زيت نخالة الأرز، الدهون القابلة للفرد، الثباتية للاكسدة، رقم البيروكسيد، رقم حامض الثيوباربتيوريك.