Linalyl Oleate as a Frying Oil Autoxidation Inhibitor

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ABSTRACT: Linalyl oleate (LO), an interesterification product of linalyl acetate (LA) and methyl oleate catalyzed with sodium methoxide, was studied to determine its effectiveness in retarding oxidative changes in soybean oil heated continuously at 180 ± 5°C for 32 h. The identity of LO was established by GC–MS and NMR. LO was tested at levels of 0.05 and 0.1% and compared with the more commonly used synthetic autoxidation inhibitor* methyl silicone (MS) at levels of 5 and 10 ppm. FA changes and conjugated dienoic acid formation were monitored. First-order kinetic equations were used to model the decreases in linoleate (18:2)/palmitate and linolenate (18:3)/palmitate ratios. Plots of the data show an inflection point at ~11 h. Oils with either level of MS and LO had lower reaction rate constants before the inflection points, and lower conjugated diene values and higher 18:2 and 18:3 percentages at the end of the 32-h heating period than did oil without additives and with LA. LO could replace methyl silicone in soybean oil during deep-fat frying but at levels about 100 times greater. [*We propose to use the term "autoxidation inhibitor" for substances that inhibit autoxidation when added to fats and oils at low concentrations and whose mechanism of action may be unknown. Some may wish to call such substances "antioxidants" but others wish to reserve this term for substances that end free radical chains by hydrogen radical donation. Some refer to methyl silicone as a "polymerization inhibitor," but this term suggests more about its mechanism of action than seems warranted.]

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During frying, fat is exposed to elevated temperatures and atmospheric oxygen, resulting in deterioration in flavor, color, and nutritive value of the oil, especially reductions in the content of EFA. The main changes occurring during frying include oxidation, hydrolysis, and polymerization.

Oxidation can be retarded by adding antioxidants, but most phenolic antioxidants undergo distillation or destruction in deep-fat frying conditions, thus minimizing their protective effect under these conditions. The commonly used synthetic autoxidation inhibitor[1] for frying oil is methyl silicone (MS) (1). MS was originally used in frying oils to prevent foaming, and its mechanism for retarding oxidation is uncertain. One hypothesis is that it accumulates in the oil surface and acts as an oxygen barrier. Disadvantages of using MS include loss of volume in cake baking, batter defoaming in doughnut frying, and loss of crispness in fried potato chips (2).

Many people prefer to have "natural" autoxidation inhibitors in their food, but so far no natural frying autoxidation inhibitor is both effective and available. A number of plant sterols, including Δ^5 - and Δ^7- avenasterol (Fig. 1), vernosterol, and citrostadienol, reduce the chemical changes that occur in vegetable oils during frying (3,4). Gordon and Magos (3) theorized that the ethylidine side chain present on these sterols reacts rapidly with lipid free radicals to form stable allylic tertiary free radicals that are too weak to continue the oxidation chain. The ethylidene side chain forms free radicals rapidly because of the presence of unhindered hydrogen atoms on an allylic carbon atom.

Linalool (Fig. 1), a terpenol compound found in herbs such as basil and coriander, contains a double bond structure similar to that found in the plant sterols and has a prooxidative effect in frying oil when present at levels above 0.05% (5). But this prooxidative effect can be avoided by esterification of the hydroxyl group of linalool, for instance with linalool acetate (LA; Fig. 1). The disadvantages of LA are that it possesses a relatively

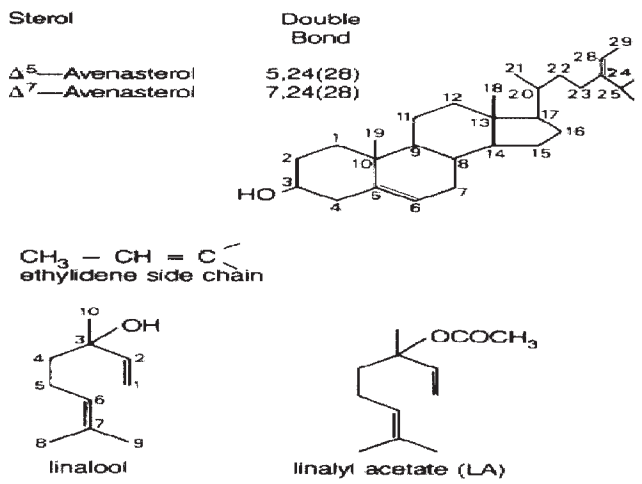

FIG. 1. Chemical structures of compounds containing an ethylidene group.

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[1]We propose to use the term "autoxidation inhibitor" for substances that inhibit autoxidation when added to fats and oils at low concentrations and whose mechanism of action may be unknown. Some may wish to call such substances "antioxidants" but others wish to reserve this term for substances that end free radical chains by hydrogen radical donation. Some refer to methyl silicone as a "polymerization inhibitor," but this term suggests more about its mechanism of action than seems warranted.

strong flavor and tends to distill out of the fat at frying temperature. By bonding the linalool to oleic acid, we increased its M.W. and made it less flavorful and volatile. The objectives of the present study were to develop a process for creating linalyl oleate (LO), to evaluate the effectiveness of LO as an autoxidation inhibitor in heated soybean oil (SBO), and to test for a possible synergistic effect of an LO–MS mixture.

EXPERIMENTAL PROCEDURES

Materials. (i) Oils. Bleached, deodorized SBO, containing only citric acid, was a gift from the Archer Daniels Midland Company (Decatur, IL). PV of the SBO as received were 0.1 meq/kg by AOCS method Cd 8-53 (6). Olive oil was purchased from a local market.

Silica gel (40–140 mesh) used for purification of the LO reaction mixture was obtained from J.T.Baker Inc. (Phillipsburg, NJ). Urea and sodium methylate solution in methanol (~5.4 M) was purchased from Fluka (Milwaukee, WI). Other chemicals were reagent grade from Fisher (Fairlawn, NJ).

(ii) Autoxidation inhibitors. Food-grade polydimethyl siloxane or MS fluid (MS: 0.97 g/mL at 25°C; viscosity, 350 centistokes; stock no. 200®) was a gift from Dow Corning Co. (Midland, MI). LA was purchased from Aldrich Chemical Co. (Milwaukee, WI).

Concentration of methyl oleate (MO). MO was concentrated from olive oil by urea fractionation (7). An extra step was introduced to remove the saturated esters, and a distillation was not performed. Olive oil (100 g) was added to 500 mL of boiled methanol. When the mixture reached the b.p., 5 mL of 5.4 M sodium methylate solution was added and the mixture was refluxed. Next an additional 500 mL methanol and 210 g urea were added. The mixture was boiled until the urea dissolved, cooled to room temperature, and left overnight. The mixture then was filtered to obtain a liquid fraction rich in MO and a crystalline fraction rich in methyl palmitate. An additional 150 g urea was added to the liquid fraction, and it was boiled and left overnight, as before. The crystalline fraction was rich in MO and poor in both methyl palmitate and methyl linoleate. The crystalline fraction was stirred with water containing several drops of concentrated hydrochloric acid (pH 3.0), and the MO concentrate was collected for synthesis of LO.

Synthesis of LO. LO was synthesized by interesterification of LA and a 10% molar excess of MO with 5 mL of 5.4 M methanolic sodium methoxide solution from which the methanol was removed under vacuum before the other reagents were added. A reduced pressure was used to remove methyl acetate and drive the reaction toward the formation of LO.

Column purification of LO. The LO mixture was fractionated by LC to remove unreacted MO. One gram of the reaction mixture was passed into a column (20 mm i.d. × 18 cm) containing 10 g of silica gel using hexane/diethyl ether (1:0.005 vol/vol). The ether was distilled over lithium aluminum hydride to remove peroxides and the BHT it contained as a stabilizer. Elution fractions of 20 mL were collected; and the fourth, fifth, and sixth fractions contained 92–95% LO by GC.

Frying procedure and oil sampling. Oil samples (200 g), with and without the various additives, were heated continuously in FryBaby® 05430 (National Presto Industries, Eau Claire, WI) deep fat fryers at 180 ± 5°C for 32 h. Autoxidation inhibitors were dissolved in distilled ethanol, and the ethanol was vaporized before the oils were added. The LO was added to SBO at levels of 0.05 and 0.1%. MS was tested at 5 and 10 ppm. The mixture of 0.05% LO and 5 ppm MS was tested for synergistic effect, and LA was tested at a concentration equimolar to 0.05% LO (~0.025%).

The temperature of each fryer was maintained by a variable transformer and monitored with a thermocouple. Three grams of oil was removed at 2, 4, 8, 12, 24, and 32 h for analyses and stored under nitrogen at 5°C until analyzed. The oil removed for samples was not replenished.

Conjugated dienoic acids (CD). CD were measured by AOCS method Ti 1a-64 (6) with a Hitachi U-2000 model spectrophotometer.

GC analyses. FAME of the frying oils were prepared by transesterifying the oils with sodium methoxide in methanol and injecting in a gas chromatograph (GC), as described by Hammond (8). An HP 5890 Series II GC equipped with an FID was used. A fused-silica capillary column (15 m × 0.25 mm × 1.0 μm film thickness, coated with SP-2330; Supelco, Bellefonte, PA) was used. Helium was the carrier gas (3 mL/min), and the injection port and detector temperatures were set at 230°C. The column was temperature programmed at 10°C/min from 140 to 225°C and maintained at 225°C for 5 min for the liquid and solid fractions obtained during the isolation of MO. For FA compositions of the heated oil samples, the column temperature was maintained at 190°C for 5 min. For the LO, the column was programmed at 10°C/min from 100 to 225°C. A MicroMass CA 062 GC-MS with a DB-23 (Agilent, Wilmington, DE) fused-silica capillary column (30 m × 0.25 mm × 0.2 μm film thickness) and an Agilent 6890 series GC were used to verify the chemical structure of LO. The injection and detection port temperatures were both 230°C. The column was held at 80°C for 1 min and raised at 20°C/min from 80 to 260°C and held for 5 min with helium as the carrier gas. The split ratio was 50%.

NMR spectra. ^{1}H NMR spectra were obtained on a Varian VXR 300-MHz instrument.

Surface tension measurement. A FACE Automatic Surface Tensiometer (Model CBVP-Z; Tantec, Schaumberg, IL) was used.

Statistical analysis. All data are the average of replicate experiments. Data from analyses were analyzed statistically using the one-way ANOVA by means of general linear models procedure of a SPSS 9.0 (Chicago, IL) software package. Surface tension data also were analyzed using a paired comparison *t*-test.

RESULTS AND DISCUSSION

The MO concentrated by urea fractionation was 83.7% MO, 9.8% methyl linoleate, and 0.4% methyl palmitate; the area

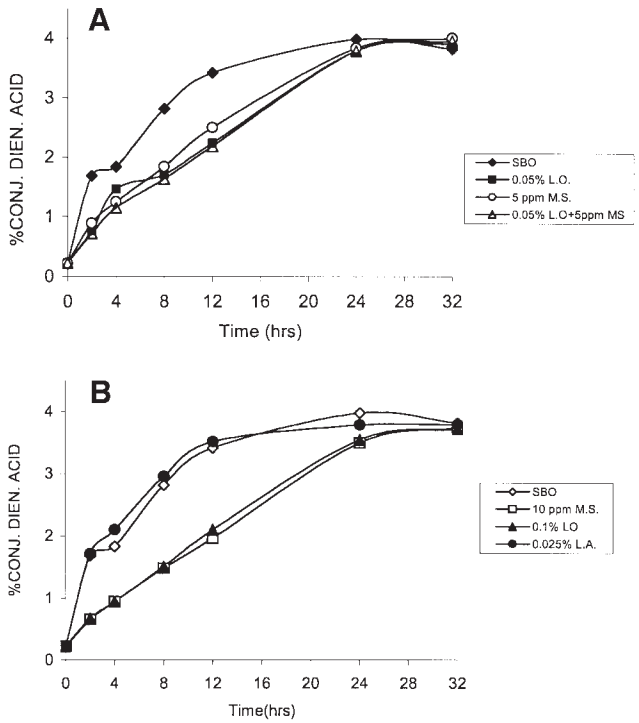

FIG. 2. Percentages of conjugated dienoic acid in soybean oil (SBO) protected with (A) 0.05% linalyl oleate (LO), 5 ppm methyl silicone (MS), and 0.05% LO + 5 ppm MS and (B) 0.1% LO, 10 ppm MS, and 0.025% linalool acetate (LA).

percentages of other peaks were lower than 1%. After the interesterification reaction of the MO and LA, the product was analyzed by GC, and it was found to be 10.4% MO, 74.9% LO, and 8.6% linalyl linoleate. Other peaks had percentages lower than 1%. After silica column chromatography, the product consisted of 0.4% MO, 1.0% linalyl palmitate, 92.3% LO, and 2.9% linalyl linoleate, and the percentages of other peaks were lower than 1%. The yield of purified linalyl ester was 33% of the material placed on the column.

GC and GC–MS to identify the chemical composition of LO. Although LO was synthesized previously by reaction of oleyl chloride with linalool (9), adequate GC–MS or NMR spectra for LO are lacking in the literature. The expected M.W. of LO is ~418.72. Chemical ionization GC–MS gave a mass of 418.39. On a polar SP2330 column, LO emerges at ~205°C, but on a nonpolar DB5 (Agilent) column it begins to decompose at 225°C before it has eluted. GC–MS revealed the primary decomposition product to be ocimene ($C_{10}H_{16}$) formed by the dehydration of linalool. Yan and White (5) reported that terpenols and their esters undergo elimination and rearrangement reactions when they are subjected to intense heat, steam distillation, and/or acidic conditions.

NMR to identify the chemical composition of LO. Chemical shifts are given in ppm (δ) and multiplicities are indicated by *s* (singlet), *d* (doublet), *t* (triplet), *q* (quartet), *qn* (quintet), and *m* (multiplet). ^{1}H NMR (300 MHZ, $CDCl_3$, 16 mg/mL) for LO: δ: 0.87 (4H, *t*), 1.26 (26H, *q*), 1.53 (12H, *q*), 1.83 (9H, *m*), 2.25 (3H, *t*), 3.47 (0.7H, *q*), 5.09 (4H, *qn*), 5.34 (3H, *qn*), 5.91 (1.5H, *q*).

CD. The percentages of CD of oil without additives (control) and oils with 0.05% LO, 5 ppm MS, and 0.05% LO + 5 ppm MS during the 32 h heating period are shown in Figure 2A. The percentage of CD of the control increased for 24 h, reached a plateau, and decreased. At the end of 32 h the control had the lowest CD percentage of all the treatments. The MS and LO additives retarded the increase in percentage of CD similarly, and at the end of 32 h their CD percentages were still increasing.

Figure 2B shows the changes in percentage of CD in control and in oils with 0.1% LO, 10 ppm MS, and 0.025% LA. MS and LO showed exactly the same decrease in the formation of percentage of CD relative to the control. The activity of all the additive treatments except LA was statistically significant through 24 h compared with the control. LA did not show any inhibitory activity and may be slightly prooxidative when compared with the control. After 24 h, the percentages of CD in the control and LA-treated oil started to decrease, whereas in oils with LO and MS they were still increasing. Control and oils with additives reached similar CD percentages at the end of 32 h.

CD formation and changes in FAME percentages have been shown to parallel polymer formation in heated oils (4,5,10). In particular, formation of CD has been shown to have a linear relationship with total polar compounds, which provide a reliable measure of the extent of deterioration (11–14).

FA composition. The FA profiles of the control and oils with additives are shown in Table 1. Significant differences in FAME percentages for each treatment are shown for each sampling period. For all treatments, the percentages of PUFA (18:2 and 18:3) tended to decrease during heating, whereas the percentages of the saturated (16:0 and 18:0) and monounsaturated (18:1) FA tended to increase. The percentages of 16:0 in samples containing all levels of LO and MS generally were significantly lower through the 32-h heating period than the control and the oil with LA ($P < 0.05$). After 4 h, the 18:0 and 18:1 percentages of oils containing all levels of LO and MS generally were significantly lower, and the percentages of 18:2 and 18:3 were significantly higher than those of the control and of oil containing LA ($P < 0.05$). Among the oils with additives, oils containing 0.1% LO and 10 ppm MS had significantly lower 16:0, 18:0, and 18:1 percentages and higher 18:2 and 18:3 percentages than oils with lower levels of LO and MS and their mixtures ($P < 0.05$).

The ratio of 18:2 and 16:0 percentages are often used as indicators of the extent of fat deterioration because linoleate esters are quite susceptible to oxidation, whereas palmitate esters are stable. This ratio has been reported to correlate with the iodine value and dielectric constant (13,15). The ratio of 18:2%/16:0% is a measure of the surviving percentage of 18:2 as frying time increases.

In Figure 3A, the natural logarithm of the 18:2%/16:0% values from Table 1 of control and oils with 0.05% LO, 5 ppm MS, and the LO + MS mixture are shown over the 32-h heating period. A linear decrease with frying time was observed for the control, and the data were fitted to first-order kinetics. Oils with additives had inflection points between 11 and 13 h, and

TABLE 1
FA Composition[a] (%) of Soybean Oil (SBO) Treatments Heated at 180 ± 5°C

Treatment	16:0	18:0	18:1	18:2	18:3
0 Hour					
SBO	10.3	4.4	25.6	52.1	7.1
2 Hours					
SBO	10.9^{b}	4.8^{b}	24.5^{c}	50.4^{a}	6.4^{a}
0.05% LO	10.6^{a}	4.5^{a}	23.7^{a}	51.4^{b,c,d}	6.9^{b}
5 ppm MS	10.6^{a}	4.6^{a,b}	24.0^{a,b,c}	51.1^{a,b,c}	6.7^{a,b}
0.05% LO + 5 ppm MS	10.5^{a}	4.7^{a,b}	23.9^{a,b,c}	51.2^{b,c,d}	6.8^{a,b}
10 ppm MS	10.4^{a}	4.5^{a}	23.7^{a}	51.8^{c,d}	6.9^{b}
0.1% LO	10.5^{a}	4.5^{a}	23.8^{a,b}	51.9^{d}	6.9^{b}
0.05% L.A.	10.9^{b}	4.7^{a,b}	24.5^{b,c}	50.7^{a,b}	6.4^{a}
4 Hours					
SBO	11.4^{b}	5.0^{a}	25.1^{a}	49.7^{a}	6.0^{a}
0.05% LO	10.7^{a}	4.8^{a}	24.4^{a}	50.7^{a,b}	6.5^{a,b}
5 ppm MS	10.8^{a}	4.8^{a}	24.3^{a}	50.6^{a,b}	6.5^{a,b}
0.05% LO + 5 ppm MS	10.7^{a}	4.8^{a}	24.3^{a}	50.6^{a,b}	6.5^{a,b}
10 ppm MS	10.6^{a}	4.6^{a}	23.9^{a}	51.4^{b}	6.7^{b}
0.1% LO	10.6^{a}	4.6^{a}	24.0^{a}	51.5^{b}	6.7^{b}
0.05% LA	11.3^{b}	4.9^{a}	25.0^{a}	49.8^{a}	6.1^{a,b}
8 Hours					
SBO	11.9^{b}	5.1^{b}	25.6^{b}	48.2^{a}	5.5^{a}
0.05% LO	10.9^{a}	4.7^{a}	24.2^{a}	50.4^{b}	6.3^{a,b}
5 ppm MS	11.0^{a}	4.7^{a}	24.4^{a}	50.2^{b}	6.2^{a,b}
0.05% LO + 5 ppm MS	10.9^{a}	4.7^{a}	24.3^{a}	50.5^{b}	6.4^{a,b}
10 ppm MS	10.8^{a}	4.7^{a}	24.5^{a}	51.0^{b}	6.4^{b}
0.1% LO	10.8^{a}	4.7^{a}	24.5^{a}	51.0^{b}	6.4^{b}
0.05% LA	12.3^{b}	5.3^{c}	26.4^{c}	48.4^{a}	5.4^{a,b}
12 Hours					
SBO	12.8^{b}	5.5^{b}	26.7^{b}	45.9^{a}	4.8^{a}
0.05% LO	11.3^{a}	4.8^{a}	24.8^{a}	49.4^{b}	5.9^{b}
5 ppm MS	11.5^{a}	4.9^{a}	25.0^{a}	49.0^{b}	5.8^{b}
0.05% LO + 5 ppm MS	11.3^{a}	4.8^{a}	24.8^{a}	49.4^{b}	5.9^{b}
10 ppm MS	11.3^{a}	4.8^{a}	25.0^{a}	50.2^{b}	6.1^{b}
0.1% LO	11.2^{a}	4.9^{a}	24.9^{a}	50.0^{b}	6.0^{b}
0.05% LA	13.3^{c}	5.7^{c}	27.6^{c}	45.8^{a}	4.6^{a}
24 Hours					
SBO	15.6^{b}	6.7^{c}	30.1^{b,c}	38.9^{a}	3.0^{a}
0.05% LO	13.9^{a}	5.9^{a,b}	27.9^{a}	44.7^{b,c}	4.0^{b,c}
5 ppm MS	14.3^{a}	6.1^{b}	28.6^{a,b}	42.3^{b}	3.7^{b}
0.05% LO + 5 ppm MS	13.9^{a}	6.0^{a,b}	28.3^{a}	43.1^{b,c}	3.9^{b,c}
10 ppm MS	13.5^{a}	5.7^{a,b}	27.9^{a}	45.4^{c}	4.4^{d}
0.1% LO	13.6^{a}	5.6^{a}	28.0^{a}	44.2^{b,c}	4.2^{c,d}
0.05% LA	16.4^{b}	6.9^{c}	31.1^{c}	38.7^{a}	2.9^{a}
32 Hours					
SBO	17.2^{d}	7.4^{d}	31.8^{c}	35.2^{a}	2.4^{a}
0.05% LO	15.6^{a,b,c}	6.7^{a,b,c}	30.3^{a,b}	38.7^{b,c,d}	3.0^{b,c,d}
5 ppm MS	16.1^{c}	6.9^{c}	30.7^{b}	37.7^{b}	2.8^{b}
0.05% LO + 5 ppm MS	16.0^{b,c}	6.8^{b,c}	30.6^{b}	38.4^{b,c}	2.9b,c
10 ppm MS	15.2^{a}	6.5^{a}	29.8^{a}	39.9^{d}	3.2^{d}
0.1% LO	15.4^{a,b}	6.7^{a,b}	30.1^{a,b}	39.4^{c,d}	3.1^{c,d}
0.05% LA	17.9^{e}	7.6^{d}	32.2^{c}	34.5^{a}	2.3^{a}

[a]FAME percentages in the same column and at same time that have the same superscript are not significantly different ($P < 0.05$).

the reaction rate before the inflection point was lower than the reaction rate after the inflection point, but both parts of the plots showed linearity.

Figure 3B also shows the same tendencies of oils with 0.1% LO and 10 ppm MS that were noted with lower concentrations of these additives. The 18:2%/16:0% in oil with LA had no inflection point.

Lines were fitted before and after the inflection points of oils with additives, resulting in the reaction rate constants shown in the Table 2. Oils with 10 ppm MS, 0.05% LO, and 0.1% LO had similar rate constants before and after the inflection points, and the reaction rates of oils with additives, other than LA, were not significantly different from each other ($P > 0.05$). Before the inflection point, all levels of LO and MS reduced the

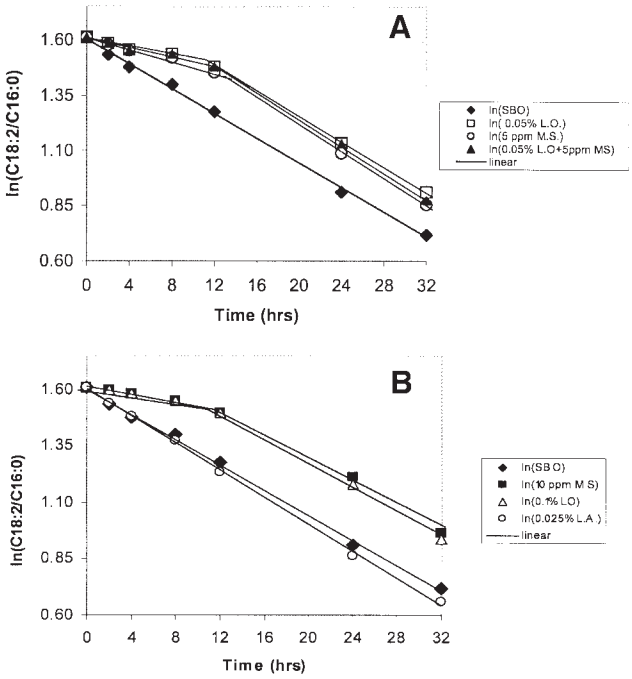

FIG. 3. Decrease of natural logarithm of 18:2/16:0 in SBO (control) and SBO protected with (A) 0.05% LO, 5 ppm MS, and 0.05% LO + 5 ppm MS and (B) 0.1% LO, 10 ppm MS, and 0.025% LA, both as a function of frying time. For abbreviations see Figure 2.

rate of disappearance of linoleate by 2.5- or 3.1-fold compared with control oil. After the inflection points, the rate constants were similar to those of the control oil. The inflection point of oil with 0.05% LO + 5 ppm MS occurred later in frying than any of the other oils, but its reaction rate was third-highest among the treatments.

Rate constants of changes in linolenate%/palmitate% are shown in Table 3. The rate constants for the control and oil with LA were 0.050/h and 0.053/h, respectively, which were greater than constants for oils with either level of LO and MS. The reaction rates of oils with additives other than LA were significantly lower than the control before the inflection points. After the inflection points, the rates were comparable to those of the control. The rate of decrease in 18:3 was about 1.8 that of comparable values for 18:2, which is close to the value of 2.0 found for ambient temperature oxidation (16).

The addition of additives, except LA, exhibited autoxidation inhibition in SBO at deep-fat frying temperature, and the additives improved the oxidative stability of the oil. Previous work reported that LA was not effective at the concentration we used (5). The effect of a LO + MS mixture for retarding oxidation of SBO was not better than either 0.05% LO or 5 ppm MS and thus showed no synergism. The results indicated that LO could replace MS in SBO during deep-fat frying but at levels about 100 times greater. In addition, the recommended level for LO is 5 times greater than that allowed for the phenolic antioxidants used for ambient temperature oxidation.

The kinetic plots of the oils treated with additives show an inflection at about 11 h after which the rate of disappearance of the polyunsaturates is similar to that of the control oil (Tables 2 and 3). Thus, the additives no longer seem to be active after about 11 h and perhaps are exhausted by the end of this period. Exhaustion of LO could support the ethylidene oxidation theory of Gordon and Magos (3), but it is not clear how MS would be exhausted and lose its inhibitory potency at about the same time as LO. Possibly, the inhibitory activity of both compounds is being controlled by some other substance that is being used up at about 11 h. If so, tocopherols are good candidates for the compounds that are exhausted at the time of the inflection point. Frankel (2) suggested that MS may act synergistically with TBHQ and other phenolic antioxidants under frying conditions.

Near the end of the 32-h heating period, we observed that oils began to form surface skins. During continued heating, the skin spread to cover more of the surface on oils containing LO or MS than on the control oil. This observation might be considered to support a surface film mechanism for the action of MS and LO (1). Although the skin was not apparent until about 32 h of heating, it could have been exerting an effect before it was observable. However, the kinetics in Tables 2 and 3 show that after the inflection points, the oils with additives had the same reaction rates as the control oil, so there seems to have been no obvious advantage that can be attributed to surface skin formation since it showed no effect when it was most obvious

TABLE 2
Rate Constants[a] of the Changes in 18:2/16:0 Ratio During Heating at 180 ± 5°C

Treatment	Before inflection point $C_{18:2}/C_{16:0}$, k (h^{-1})	Inflection point (h)	After inflection point $C_{18:2}/C_{16:0}$, k (h^{-1})
Soybean oil	–0.028[b]	—	–0.028[a,b]
0.05% LO	–0.009[a]	11.1	–0.028[a,b]
5 ppm MS	–0.012[a]	11.2	–0.030[b]
0.05% LO + 5 ppm MS	–0.011[a]	12.0	–0.030[b]
10 ppm MS	–0.009[a]	11.6	–0.026[a]
0.1% LO	–0.008[a]	11.2	–0.028[a,b]
0.025% LA	–0.030[b]	—	–0.030[b]

[a]Constants within a column with the same superscript are not significantly different ($P < 0.05$). For abbreviations see Table 1.

TABLE 3
Rate Constants[a] of the Changes in 18:3/16:0 Ratio During Heating at 180 ± 5°C

Treatment	Before inflection point $C_{18:3}/C_{16:0}$, k (h^{-1})	Inflection point (h)	After inflection point $C_{18:3}/C_{16:0}$, k (h^{-1})
Soybean oil	–0.050[b]	—	–0.050[a,b,c]
0.05% LO	–0.021[a]	11.9	–0.051[a,b,c]
5 ppm MS	–0.024[a]	11.7	–0.054[b,c]
0.05% LO + 5 ppm MS	–0.023[a]	11.8	–0.054[c]
10 ppm MS	–0.020[a]	12.3	–0.047[a]
0.1% LO	–0.019[a]	12.0	–0.049[a,b]
0.025% LA	–0.053[b]	—	–0.053[b,c]

[a]Constants within a column with the same superscript are not significantly different ($P < 0.05$). For abbreviations see Table 1.

and widespread. Thus, the eventual formation of a surface skin does not seem to be the mechanism by which LO and MS slow the oxidative changes in the oil. The skin was recovered by placing a metal screen under the surface and lifting out the skin. The skin was washed with hexane and converted to methyl esters. Analysis of the esters (data not shown) revealed elevations of saturates and decreases of polyunsaturates that were greater in extent than those observed for the bulk oils in Table 1. There also were peaks with longer retention times that probably represent oxidized esters that were not observed in the bulk oil.

The surface tension at ambient temperature of the SBO was 33.3 mN/m. The addition of MS in concentrations varying from 1 to 10 ppm in 1 ppm steps and LO ranging from 0.01 to 0.1% in 0.01% steps gave surface tensions ranging from 30.8 to 32.9 and 32.8 to 33.3 mN/m, respectively. Duplicates for MS showed considerably more variation than those of the control and LO additions. None of the levels of addition of LO or MS were significantly different from the control oil in a paired comparison test at $P \leq 0.05$, and there was no convincing trend for the surface tension to decrease with the amount of MS or LO added. But most of the additions of LO and MS gave slightly lower surface tensions than the control oil, and comparisons of the levels of MS and LO as groups with the controls were significantly different for both additives. However, these observations do not make a convincing case for the accumulation of MS or LO in the oil surface.

REFERENCES

1. Gordon, M.H., The Mechanism of the Antioxidant Action *in vitro,* in *Food Antioxidant,* edited by B.F.J. Hudson, Elsevier, New York, 1990, pp. 13–14.
2. Frankel E.N., *Lipid Oxidation*, Oily Press, Dundee, Scotland, 1998, pp. 244–245.
3. Gordon, M.H., and P. Magos, The Effect of Sterols on the Oxidation of Edible Oils, *Food Chem. 10*:141–147 (1983).
4. White. P.J., and L.S. Armstrong, Effect of Selected Oat Sterols on the Deterioration of Heated Soybean Oil, *J. Am. Oil Chem. Soc. 63*:525–529 (1986).
5. Yan, P.S., and P.J. White, Linalyl Acetate and Other Compounds with Related Structures as Antioxidants in Heated Soybean Oil, *J. Agric. Food Chem. 38*:1904–1908 (1990).
6. *Official Methods and Recommended Practices of the American Oil Chemists' Society*, 4th edn., edited by D. Firestone, AOCS Press, Champaign, 1989.
7. Swern, D., Techniques of Separation*,* in *Fatty Acids, Their Chemistry, Properties, Production, and Uses*, edited by K.S. Markley, Interscience Publishers, New York, 1964, pp. 2328–2329.
8. Hammond, E.G., Rapid Analysis of Lipids in Many Individual Plants, in *Modern Methods of Plant Analysis*, New Series, Vol. 12, edited by H.F. Liskens and J.F. Jackson, Springer-Verlag, New York, 1991, pp. 321–330.
9. Van Duuren, B.L., T. Blazej, B.M. Goldschmidt, C. Katz, S. Melchionne, and A. Sivak, Cocarcinogenesis Studies on Mouse Skin and Inhibition of Tumor Induction, *J. Natl. Cancer Inst. 46*:1039–1044 (1971).
10. White, P..J., and Y.-C. Wang, A High Performance Size-Exclusion Chromatographic Method for Evaluating Heated Oils, *J. Am. Oil Chem. Soc. 63*:914–920 (1986).
11. Fritsch, C.W., Measurements of Frying Fat Deterioration: A Brief Review, *Ibid. 58*:272–274 (1981).
12. Richard, E., F. Stier, and M. Blumenthal, Quality Control in Deep–Fat Frying, *Baking Snack 15*:67–76 (1993).
13. Houhoula, D.P., V. Oreopoulou, and C. Tzia, A Kinetic Study of Oil Deterioration During Frying and a Comparison with Heating, *J. Am. Oil Chem. Soc. 79*:133–137 (2002).
14. White. P.J., Methods for Measuring Changes in Deep–Fat Frying Oils, *Food Technol. 45(2)*:75–80 (1991).
15. Augustin, M.A., T. Asap, and L.K. Heng, Relationships Between Measurements of Fat Deterioration During Heating and Frying in RBD Olein, *Ibid. 64*:1670–1675 (1987).
16. Fatemi, S.H., and E.G. Hammond, Analysis of Oleate, Linoleate and Linolenate Hydroperoxides in Oxidized Ester Mixtures, *Lipids 15*:379–385 (1980).

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