CrossMark

Effect of antioxidant active films on the oxidation of soybean oil monitored by Fourier transform infrared spectroscopy

Pablo S. Anbinder [a,b], Pablo J. Peruzzo [a,b], Miriam N. Martino [b,c], Javier I. Amalvy [a,d,e,f,*]

[a] Instituto de Investigaciones Fisicoquímicas Teóricas y Aplicadas (INIFTA), CCT La Plata CONICET-UNLP, Diag. 113 y 64, La Plata, Argentina
[b] Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina
[c] Centro de Investigación y Desarrollo en Criotecnología de Alimentos (CIDCA, CCT La Plata CONICET-UNLP), Calle 47 y 115, La Plata, Argentina
[d] Comisión de Investigaciones Científicas de la Provincia de Buenos Aires (CICPBA), Argentina
[e] Cátedra de Materiales Poliméricos de la Facultad de Ingeniería, Universidad Nacional de La Plata, Calle 1 y 47, La Plata, Argentina
[f] Centro de Investigación y Desarrollo en Tecnología de Pinturas (CIDEPINT, CIC-CCT La Plata CONICET), Av. 52 entre 121 y 122, La Plata, Argentina

ARTICLE INFO

ABSTRACT

An active film prepared from waterborne polyurethane incorporated with different concentrations of butylated hydroxytoluene (BHT) and α-tocopherol was developed, and the films' antioxidant characteristics were studied using soybean oil as food model. The release behavior of additives from such films was studied using isooctane as a simulant fatty food. Antioxidant activity of the films into soybean oil at three experimental conditions was monitored by FTIR spectroscopy. Induction time and the kinetics of the oxidation process were followed using different bands of the FT-IR spectra. Films incorporated with 2 wt.% of BHT or α-tocopherol showed enough antioxidant activity to preclude oil oxidation during 60 days at 60 °C in the dark. The results showed that this type of active films could be used for food preservation as part of a packaging containing oils or oily related products.

© 2014 Elsevier Ltd. All rights reserved.

1. Introduction

The active packaging is one of the most novel concepts that has arisen in the food package industry, answering to the continuous demands of the consumers for more fresh products and of the producers with extended shelf life (Vermeiren et al. 1999; Brody et al., 2001; Ahvenainen, 2003).

Beneficial interactions between food and package are based on the regulation of the headspace content (O_2, CO_2, ethylene), the control of humidity, the action of diverse enzymes, liberation of antimicrobial or antioxidant substances, among others. This approach can reduce the addition of large amounts of food additives that are usually incorporated into food.

Originally, inside the package only physicochemical changes occurred due to the natural evolution of the product, with this new technology the headspace environment will be improved owing to the package (Rooney, 2005).

Oxidation of lipid compounds in the packaged foods could be controlled by the use of antioxidant additives. If antioxidants are included in the matrix of the packing material and a controlled slow liberation is achieved, the contained product will see its shelf life increased, without adding unnecessary quantities of the antioxidant. Synthetic antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) although very effective, are not without side-effects (Shlian and Goldstone, 1986) and therefore it should be limited by regulating the dosage (controlled release for example) or by substituting them with natural antioxidants like tocopherols.

Oils and fat oxidation is one of the most noticeable degradation processes in food industry, due to the generation of undesirable flavors and changes in nutritional aspects, with consequences on health, nutritional quality and consumer's choice (Gómez-Alonso et al., 2004). During lipid degradation, different processes occur simultaneously: oxidation of some compounds, hydrolysis of triglycerides and *cis/trans* isomerization of unsaturated fatty acids (Li et al., 2013; Velasco and Dobarganes, 2002).

The lipid degradation process occurs at low rate at room temperature, and hence accelerated methods should be employed to estimate the oxidative stability of the product or the induction time of the autoxidation reaction in a relatively short period of time (Frankel, 1993). Several physical or chemical parameters, such as temperature can be used to increase the rate of the reaction and, consequently, the development of rancidity. In the literature,

* Corresponding author at: Instituto de Investigaciones Fisicoquímicas Teóricas y Aplicadas (INIFTA), CCT La Plata CONICET-UNLP, Diag. 113 y 64, La Plata, Argentina. Tel.: +54 221 4257291/4257430x154; fax: +54 221 4254642.
E-mail addresses: jamalvy@inifta.unlp.edu.ar, jamalvy@gmail.com (J.I. Amalvy).

several approaches to predict oil oxidative stability can be found depending on the interest in storage stability or processing performance. Oils are treated at 100 °C or lower to evaluate storage stability; in contrast, higher temperatures are used to evaluate oil performance in food processing such as frying (Climaco-Pinto et al., 2010; Guillén and Cabo, 1999; Innawong et al., 2004; Moros et al., 2009; Moya-Moreno et al., 1999; Vlachos et al., 2006).

Infrared spectroscopy has been proven to be an important tool in the assessment of the quality and composition of various edible oils, as well as in the monitoring processes in the food industry due to low cost, excellent performance and ease of use compared with other methods (Maggio et al., 2009; Van de Voort, 1992). Bands in the infrared spectrum have been reported for edible oils in different studies (Guillén and Cabo, 1999, 2002; Innawong et al., 2004; Lerma-García et al., 2010). The studies also describe how they are modified through the process of oxidation. Following these changes in the spectra, a qualitative and quantitative determination of the progress of oxidation is possible.

BHT-impregnated film has shown that retards lipid oxidation of a packaged oatmeal cereal through its migration from the product via an evaporation/sorption mechanism (Lee et al., 2004). BHT is a small molecule that exhibits high mobility and therefore has a tendency to migrate rapidly from packaging materials into foods (Wessling et al., 1998). Slow release of BHT from the package will help extend the shelf-life of the food product. The use of antioxidant packaging has also been proposed to reduce lipid oxidation in milk (Granda-Restrepo et al., 2009).

Polyurethanes (PUs) are versatile polymers prepared from condensation of polyisocyanates and polyols. The properties of these raw materials and their ratio, expressed as NCO/OH, determine the final properties. PU based materials were not used for contact in foods probably due to migration of different components and additives used during their manufacturing. However, several polymeric materials were tested by Gramiccioni et al. (1986) using olive oil and isooctane as a liquid simulant for fatty foods and they found that the global migration values obtained with olive oil and isooctane for polyurethane were much lower than those for the PVC based materials. This situation is currently changing as a consequence of gained knowledge on PU synthesis and the availability of new raw materials. General properties of polyurethanes can be modified without using plasticizers or additives just selecting the raw materials, reducing in this way the global migration and allowing them to be used in contact with food. The urethane moiety is a suitable functional group to promote interactions with other molecules and in particular with active principles capable of forming hydrogen bonds, modifying in this way their releasing behavior. On the other hand, polyolefin polymers like polyethylene, used frequently in food industry, do not have this possibility.

Bearing in mind all these facts, in this work, polyurethane films were added with different concentrations of BHT or α-tocopherol and immersed in soybean oil. The progress of the oxidation under different conditions was evaluated by FT-IR/ATR spectroscopy. The kinetics of the antioxidant release from the polyurethane films was also studied using isooctane as liquid simulant for fatty foods.

2. Materials and methods

2.1. Active films preparation

Polymer films were prepared from a waterborne polyurethane dispersion synthesized in our laboratory as described in previous papers (Pardini and Amalvy, 2008; Peruzzo et al., 2010). The mechanical and thermal properties of the polyurethane matrix were also discussed in the above mentioned works. Incorporation of the active agent to the polymeric matrix was made by dissolving

the antioxidant compound in the minimum amount of acetone and by adding this solution, drop by drop, to the polymer dispersion under magnetic stirring at room temperature for about 10 min. The dispersion was then placed on a Teflon® plate for casting. Evaporation of water and acetone was made at room temperature and in the darkness; this process took about 24 h. The formed film was cured at 60° C for 48 h to ensure the complete coalescence of the polymer.

Samples containing 1, 2 and 5 wt.% on final film of antioxidant compound were prepared.

2.2. Oxidation studies

Twenty-five mL of commercial soybean oil were placed in glass bottles of 50 mL with an open surface area of 12 cm^2 in a dry place. The oil samples were exposed to air and active films of 3.5 cm diameter and 200 μm thickness containing 1, 2 or 5 wt.% of the active component (BHT or α-tocopherol) were immersed in the oil samples. A control sample (PU film without antioxidant) was also included and experiments were run by triplicate.

It has been shown that oxidation of vegetable oils is promoted by light, temperature and oxygen availability (Pignitter et al., 2014). In order to evaluate how these factors affect the oxidation degree, samples were kept opened under three different environmental conditions at RH 60–70%:

1. Dark place at 25 °C.
2. In a transparent glass bottle (25 °C) exposed to ambient room light at day time for 10–12 h/day.
3. Dark place at 60 °C.

2.3. FT-IR spectra acquisition and data analysis

Periodically, with previous agitation of the bottle, the infrared spectrum of the soybean oil was collected. Spectra were acquired against air reference using a Spectrum One spectrophotometer (Perkin Elmer, USA) equipped with a KBr beam-splitter and DTGS detector. A 100 μL oil drop was deposited on the diamond single reflection ATR accessory. The wavenumber range measured was 4000–650 cm^{-1} with 4 cm^{-1} resolution and 32 scans accumulated. EZ OMNIC 7.4.127 software (Thermo Electron Corporation) was used for data treatment. The ATR crystal and its surroundings were thoroughly cleaned between samples.

Vibrational spectra of oils were compared to the control zero time oil. The appearance or modification of certain bands allows determining the development of oxidative process.

2.4. Determination of the induction time by the Quality Control tool

With the purpose of avoiding experimental errors in the determination of the moment where the spectra begin to change as compared to the "zero time" sample, Quality Control tool of the EZ Omnic7.4.127 software was used. This tool allows a rapid determination of induction times without processing spectra. The fingerprint region (1500–650 cm^{-1}) of the IR spectra was used to compare the fresh oil with the other samples. The critical value of correspondence was 95%.

2.5. Determination of kinetic parameters from infrared data

Kinetic parameters and induction times were calculated using the values of absorbance of different bands associated to species formed during the oxidation processes. At moderate temperatures, the lipid autoxidation follows a pseudo-first-order kinetic, independent of the oxygen pressure (Velasco and Dobarganes, 2002; Frankel, 1998; Román-Falcó et al., 2012). If other oxidation

processes follow the same kinetic order, pseudo-first-order coefficient (k) and induction time (t_{ind}) could be easily calculated, using the method given by Román-Falcó et al. (2012). There, specific band signals in the infrared spectra are related to concentration of species by means of the Beer–Bouguer–Lambert law using the following expression:

$$\ln(S_t) = \ln(S_0) - k(t - t_{int}) \tag{1}$$

where S_t and S_0 are the reactant signal at time t and the apparent initial signal obtained from the straight line when $\ln(S_t)$ is plotted versus time. From the slope of the adjusted line the pseudo-first-order coefficient k is obtained and the induction time (t_{ind}) could be calculated by:

$$t_{ind} = \frac{\ln(S_t) - \ln(S_0)}{-k} \tag{2}$$

Another expression is used when an increase in the absorbance is observed and the signal reaches a plateau in the signal:

$$\ln\left(\frac{S_\infty - S_t}{S_\infty}\right) = -k(t - t_{ind}) \tag{3}$$

where S_∞ is the product signal at time infinite.

2.6. Antioxidant release from active films

In order to study the availability of the antioxidant to prevent oil oxidation, the release behavior of films in an experimental setup (equal films dimensions and liquid volume) similar to that used for the oxidation tests, but using isooctane as a simulant oil substrate was determined (EC, 2007). The release of the additives was followed by using UV spectroscopy at 282 nm for BHT at 287 nm for α-tocopherol.

Fick's second law was used to determine the diffusion coefficient (D), according to the Eq. (4), presented by Ritger and Peppas (1987). Also, exponential factor n in Eq. (5), which is indicative of the transport mechanism, was determined.

$$\frac{M_t}{M_\infty} = 4\left(\frac{Dt}{\pi l^2}\right)^{1/2} \tag{4}$$

$$\frac{M_t}{M_\infty} = kt^n \tag{5}$$

3. Results and discussion

3.1. Effect of additive concentration

As expected, for all conditions, the control oil sample (film with no-additive) was the most affected by the oxidizing atmosphere. The soybean oil with the 1 wt.% antioxidant film showed an improvement in oil stability, and the protective effect was more evidenced in the films containing 2 and 5 wt.% of additive, with no significant differences among these concentrations in both cases (BHT and α-tocopherol). Under the studied conditions, for each additive, a 2 wt.% concentration in the film was enough to provide the best antioxidant effect. Unless otherwise stated, the results for the films are expressed with this percentage of additive.

3.2. Induction time using the Quality Control tool

As seen in Fig. 1, when the oil samples were stored in the darkness and at 25 °C, the soybean oil control sample and the oil in which the films loaded with both BHT and α-tocopherol were immersed, did not show differences compared with the fresh oil up to about 95 days. These three samples were not significantly
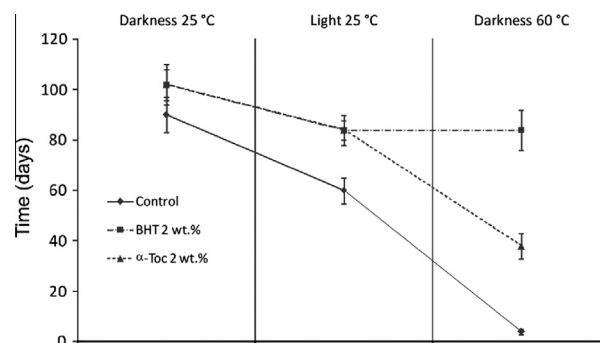

Fig. 1. Induction time of the oxidation process for samples in different storage conditions.

different from each other. The control sample showed differences with the fresh oil 60 days after the bottles had been kept at 25 °C and exposed to the day-light. Active films samples extended this period of time up to about 80 days with no differences between BHT and α-tocopherol containing films. Detectable changes were obtained in the control sample after 4 days storage at 60 °C. α-tocopherol active film retarded onset of changes in the infrared spectrum to 40 days. Oil samples in which BHT loaded films were immersed do not present changes in the fingerprint region of the IR spectra up to about 80 days under darkness conditions and at 60 °C.

Unless otherwise stated the discussion is based on results of condition 3 due to the adequate times for kinetics analysis.

3.3. Analysis of infrared data

Table 1 shows Infrared band assignments and modifications observed throughout oxidation process.

When a compound in a mixture is at a very low concentration, unless it has a very characteristic band of high absorptivity, it will produce a very weak band in the IR spectrum and will be hardly detectable. This phenomenon is observed in the band associated with OH stretching vibration of the hydroperoxide group, located around 3444 cm^{-1}, which is not observable in non-oxidized oil samples, but it can be seen as increases in intensity as oxidative process progresses (Innawong et al., 2004). This band should not be confused with the band present around 3470 cm^{-1}, assigned to an overtone of the glyceride ester carbonyl absorption (Guillén and Cabo, 2002).

Fig. 2 shows the changes in the percentage of transmittance ($T\%$) produced in the region from 3600 to 3200 cm^{-1} in the control sample for the condition 3 (dark, 60 °C). It is possible to see how the peak grows and the wavenumber shifts to lower values due to the appearance of hydroperoxide groups. In addition to the increase of the area, this band suffers deformations overtime. In the last stages, the appearance of new bands (approximately at 3530 cm^{-1}) attributed to alcohols and other secondary products of oxidation are observed along the decreasing of the hydroperoxides concentration and the shift towards the band to values of wavenumber near to those of the non-oxidized sample. Guillén and Cabo (1999) assigned different stages of the oxidation process of olive oil to the changes in this band. Briefly, the first stage (FS) was related to the period of time in which the sample is under oxidative conditions, and this spectral region remains unaltered. The second stage (SS) is related to the time interval in which hydroperoxides or primary oxidation products are being formed in appreciable proportions and so, the wavenumber of the maximum absorbance of the band begins to change from the original values to lower ones. Finally, in the third stage (TS) the concentration of

Table 1
Infrared band assignments and modifications observed throughout oxidation process.

Experimental wavenumber (cm^{-1})	Functional group	Main vibration	Wavenumber variation	Bandwidth variation	Peak absorbance variation
3560–3360	O—H hydroperoxides, alcohols, and other secondary oxidation products	O—H ν	Variable	Increase	Increase
3009	cis double bond of FA	C—H ν	No change	No change	Decrease
2923	Aliphatic (CH$_3$)	C—H asymmetrical ν	No change	No change	No change
2854	Aliphatic (CH$_2$)	C—H symmetric ν	No change	No change	No change
1800–1700	Carbonyl compounds	C=O ν	Decrease	Increase	No change
987	trans–trans conjugated dienic system	C—H δ	No change	No change	After an increase, decrease
982	cis–trans conjugated dienic system	C—H δ	No change	No change	After an increase, decrease
967	trans isolated olefins	C—H δ	Increase	Variable	Increase
914	cis double bond of FA	C—H δ	No change	No change	Decrease
723	Aliphatic (CH$_2$)	Overlapping CH$_2$ ρ and out-of-plane δ of cis disubstituted olefins	Slight increase	Decrease	No change

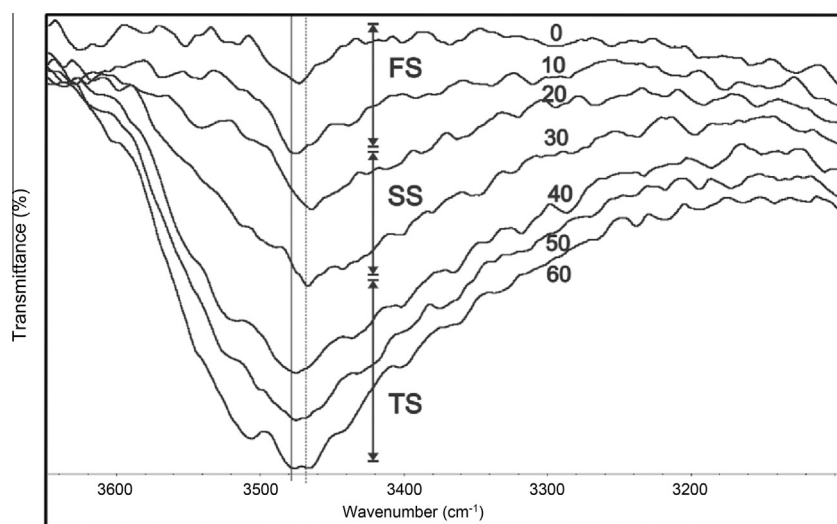ν = stretching, δ = bending, ρ = rocking.

Fig. 2. FTIR-ATR spectra in the 3600–3200 cm^{-1} region of control sample throughout the oxidation process.

hydroperoxides in the sample begins decreasing with the consequent shifting of the wavenumber of the absorbance maximum of the band back to 3444 cm^{-1}.

Control sample shows FS values of 17 days, SS between 18 and 39 days and TS from 40 days. In oil sample with α-tocopherol loaded film, no changes in wavenumber of the band are observed despite the increase in the intensity of the band. In the BHT containing sample, no increase of the band is observed, highlighting the excellent ability of this additive as primary antioxidant.

The band located at 1743 cm^{-1} was assigned to the stretching vibration of C=O ester triglyceride. This band shows a decrease in its wavenumber and a moderate broadening during the oxidation process due to the formation of secondary products (such as aldehydes and ketones). These products have bands at approximately 1728 cm^{-1}, which overlap with the bands of the ester causing those features (Guillén and Cabo, 2002).

As shifts in this band are related to degradation processes involving secondary products of oxidation, a longer induction time is expected. Fig. 3 shows t_{ind} of about 9 days in control sample and almost 50 days in α-Toc 2 wt.% sample. In BHT sample analysis the integration area of the band was not modified significantly.

Fig. 3 shows that in the control oil sample the wavenumber of the maximum of the band decreases rapidly, while oil samples
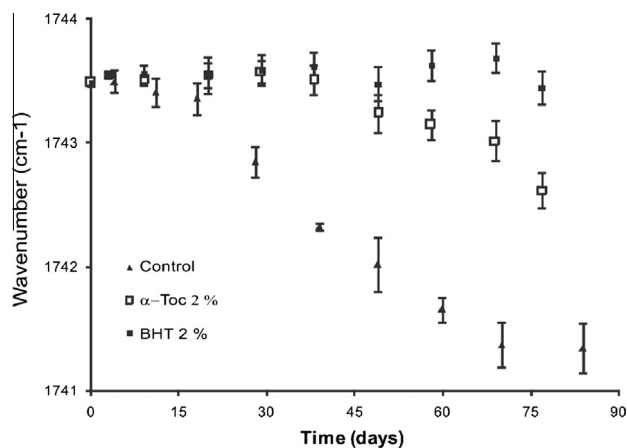

Fig. 3. Changes in the carbonyl ester band wavenumber throughout the oxidation process. Error bars indicated standard deviation (SD).

with α-tocopherol film do not show changes in this value up to 45 days. Samples with BHT film did not show changes in the wavenumber of this band for at least three months.

The wavenumber of the band assigned to the *cis* double bond (C=C—H) will vary with the oil fatty acids pattern. For soybean oil, the band attributed to this functional group is centered at 3009 cm^{-1}, while for extra virgin olive oil this band is located at 3006 cm^{-1} (Vlachos et al., 2006). The band at 914 cm^{-1} provides similar information to the band at 3009 cm^{-1} but with lower intensity, so it is less used to assess oxidative processes.

During the oxidation process, a decrease in the amount of *cis* double bonds, either for reaction (saturation of fatty acids) or by isomerization to the *trans* isomer, is observed. Thus, infrared spectroscopy allows following the fatty acids pattern during the oxidation process. Mainly two sections of the spectrum are useful for this purpose. The first one is the located at approximately 3009 cm^{-1} corresponding, as stated above, to the CH stretching band of the *cis* double bond (Vlachos et al., 2006) and the second one, through the set of bands between 930 and 995 cm^{-1} corresponding to vibrations of the double bonds of the fatty acids with *trans* configuration. Isolated *trans* double bond absorbs in the infra-

red region at a wavenumber of 967 cm^{-1} as a result of deformation out of plane of the adjacent CH bonds (Sherazi et al., 2009). When the sample exhibits a conjugated double bond system (i.e. CLA, conjugated linoleic acid), at approximately 987, 982 and 946 cm^{-1} three specific absorption peaks were recorded from the conjugated *trans–trans, cis/trans* or *trans/cis* bond, respectively (Kadamne et al., 2009).

Fig. 4 shows the temporal evolution in the control sample set of bands corresponding to the configuration of *trans* fatty acids. The band corresponding to conjugated *trans–trans* bonds (987 cm^{-1}) resulted from a free radical chain reaction and/or an intramolecular [1,3]-sigmatropic rearrangement during the linoleic oil heat treatment (Destaillats and Angers, 2005) and accumulation of this isomer, which is more stable than the *cis–cis* or *cis–trans* configuration. After having a maximum value at 30 days, this band began to fade probably due to subsequent oxidation (saturation, degradation) or by volatilization, since oxidation products are volatile and tend to escape from the oil (Climaco-Pinto et al., 2010)
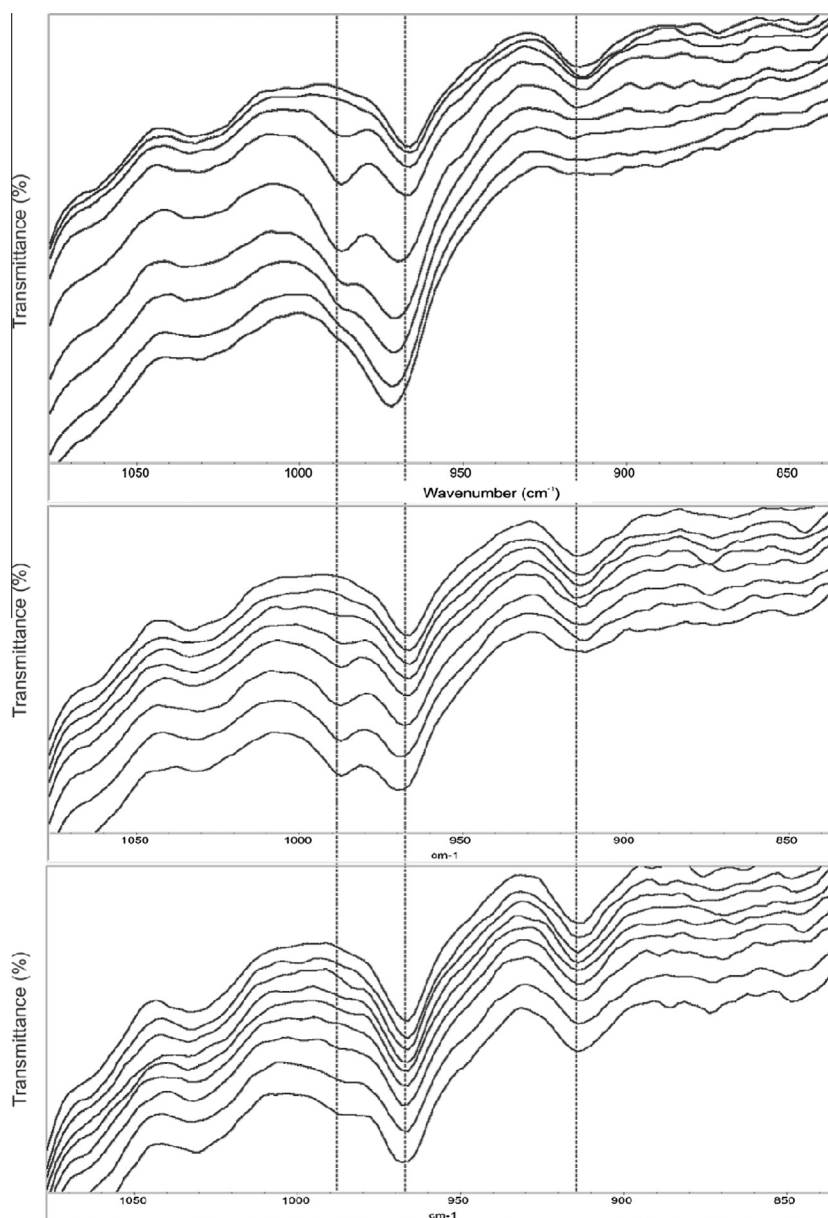

Fig. 4. Temporal evolution in the control sample (upper), α-Toc 2 wt.% (middle) and BHT 2 wt.% (bottom) for the set of bands corresponding to the configuration of *trans* fatty acids.

although the latter option is less probable at test temperature (60 °C).

The band originally located at 967 cm^{-1} appeared at high wavenumbers and became more intense throughout the oxidation process; the shift in the wavenumbers up to 973 cm^{-1} could be assigned to the accumulation of secondary oxidation products such as aldehydes or ketones supporting isolated *trans*-double bonds (Guillén and Cabo, 1999). The results reveal that over the oxidation process *cis* double bonds are transformed to *trans* isomer and finally saturated compounds are formed.

The modification of these bands was the first change in the oxidation experiment observable in the fingerprint region of infrared spectrum, and was due to an isomerization process. Same results were obtained by Guillén and Cabo (1999) in samples with a significant proportion of linolenic acyl groups such as walnut, rapeseed, and soybean oils.

Fig. 4 also shows that the intensity of the band assigned to the *cis* double bond at 914 cm^{-1} decreases.

Although the change in the spectrum of sample BHT 2 wt.% was not enough to produce a significant difference compared with the fresh oil sample, spectrum of the BHT 2 wt.% at 20 days revealed a shoulder at 983 cm^{-1} (CLA *cis–trans* diene), that was not observed in the control sample, probably due to the rate of change in *cis–trans* to *trans–trans* configuration. In light-exposed samples (room temperature) the appearance of this shoulder was also observed in the control sample.

3.4. Determination of kinetic parameters from infrared data

Table 2 shows the values of the kinetic coefficients (k) and the induction times (t_{ind}) calculated from Eqs. (2) and (3) for the different bands in the infrared spectra related to oxidative processes.

The disappearance of the peak at 3009 cm^{-1} is explained by a pseudo-first-order kinetic both in the control sample, and in the α-Toc 2 wt.%. In sample BHT 2 wt.%, although there was a trend, a very high dispersion of values occurred. Using Eq. (2), induction times of 1.5 days and of about 24 days were obtained for the sample with the control film and for the sample film with α-tocopherol, respectively. In the sample of the film containing BHT as additive the induction time was 42 days and the constant k was one order of magnitude smaller than that in the other samples.

The increase in the height of the peak located at 987 cm^{-1}, assigned to *trans–trans* double bonds conjugated system, followed a pseudo-first-order kinetics, with high values of the coefficient of determination ($R^2 > 0.98$).

In the control sample, linearity was achieved in a relatively short period of time (4–24 days). The observed induction time for the appearance of this band was 2.6 days in the control sample, 14.6 for the sample where the film was α-Toc and 18.5 days for sample with BHT active film.

It is interesting to note that while t_{ind} value was similar for samples with BHT and α-Toc, k coefficient, related to the kinetics of the process, was much lower in the first case, indicating a slow oxidative process of the oil.

The band assigned to the C=O bond showed, among those studied in this case, the largest induction time, with values of 9 days for the control sample and 49 days for the α-Toc sample, and no t_{ind} value for BHT sample at the assayed times. This result was expected, since molecules that modify the carbonyl band are secondary oxidation products.

In the control sample exposed to daylight and at 25 °C, the band at 987 cm^{-1} showed an induction time of 78 days, being the only band that could be used to perform a kinetic at the studied times.

Interesting enough is the fact that BHT containing films were more effective than α-tocopherol ones. It could be associated with a synergic effect of BHT with α-tocopherol (Marteau et al., 2014),

Table 2
Kinetic coefficients (k) and induction times (t_{ind}) of oxidative processes using associated functional groups.

Associated functional group (wavenumbers, cm^{-1})	Sample	k	t_{ind} (days)
C=C *cis* (3009)	Control	−0.01	1.5
	α-Toc 2%	−0.061	24
	BHT 2%	−0.00147	42
C=C *trans* (987)	Control	0.023	4
	α-Toc 2%	0.008	16
	BHT 2%	0.0018	20
C=O (1810–1670)	Control	0.122	8.9
	α-Toc 2%	0.0047	49
	BHT 2%	n/d	n/d

n/d: not determined.

since soybean oil contains naturally 75–120 ppm of α-tocopherol, and around 1000 ppm of total tocopherols (α-, γ- and δ-) (Ahmed et al., 2005).

3.5. Antioxidant release from active films

The migration of additives from the polymer matrix to the medium depends on, among other things, the affinity of the solute with the solvent, the matrix, temperature, characteristics of the film (pore size, free volume, T_g) and the additive (molecular size).

Table 3 shows the diffusion coefficient (D) values and the exponent n, indicative of the release mechanism of the additive, calculated using the equations (Eqs. (4) and (5)) reported by Ritger and Peppas (1987).

The D coefficient increases with increasing percentage of additive incorporation. In the α-Toc series, this increase could be related to an arrangement in small aggregates due to the limited compatibility between the additive and the polymer. α-tocopherol phytyl chain and relatively large molecular size of the α-Toc do not bind strongly to the mainly polar polymer chains under study, but they are "physically trapped" in the matrix below a critical concentration. When a greater percentage is incorporated, the size and/or possibility of connection between these aggregates will be increased and thus the migration rate will be higher. In films loaded with BHT, no significant differences were observed by varying the amount of additive incorporated. The high value of D observed for α-Toc 5 wt.% films was due to the significant surface segregation of the additive, as observed in SEM images (see Supplementary data, Fig. S2) and for that reason this sample was excluded from the systematic analysis.

Similar values of D were reported for BHT for other types of polymer (Chang et al., 1982; Schwope et al., 1987). For α-tocopherol the values found in this work were lower than those reported in other polymeric films (Zhu et al., 2012; Koontz et al., 2010). The greater rate of diffusion of BHT respect to those of α-tocopherol is also related to the smaller size of the first antioxidant molecule. This feature makes the molecules diffuse more rapidly through the polymer matrix. The effectiveness of BHT as antioxidant for certain foods was probably due in good part to its high solubility in fats and its extremely low solubility in aqueous environments

Table 3
Diffusion coefficients (D) and n values for active films in fat simulant.

Sample	D ($\times 10^{13}$) (cm^2/s)	n
BHT 1%	7.45	0.80
BHT 2%	7.59	0.80
BHT 5%	8.64	0.72
α-Toc 1%	0.79	0.93
α-Toc 2%	1.33	0.73
α-Toc 5%	31.5[a]	0.61

[a] See text for discussion.

(Phipps, 1973). Wessling et al. (1998) also found that the migration of α-tocopherol in food simulants was slower than that of BHT.

The BHT release in isooctane from the PU film should be almost complete because it is not limited by solubility. Although solubility data for BHT in isooctane is not available, this additive has good solubility in organic solvents (Schwope et al., 1987), much higher than the total amount of BHT incorporated into the film. However, as observed in previous works (Anbinder, 2011), BHT interaction with the polymer form hydrogen bonds between the phenolic OH groups of BHT molecules and the nitrogen of the urethane groups, —N(H)—C(O)O—, of polyurethane (see Supplementary material), affecting the total release of the additive (Bart, 2005). The effect of the molecular size on the rate of diffusion of these additives was also reported by Miltz and co-workers (1988) in HDPE. The mechanism that governs the release of the additives in all cases is anomalous according to Ritger and Peppas (1987) with values of n between 0.5 and 1. When the additive was BHT the value of the exponent n was not modified by their concentration. Furthermore, in films loaded with α-tocopherol, the value of n depends on the concentration. In these films the release mechanism has a diffusive contribution and dependent relaxation of the chains. Diffusive contribution becomes more important when the molecules are more likely to pass through the film as in α-Toc 5 wt.% while, in α-Toc 2 wt.%, and even more in α-Toc 1 wt.%, the release process requires polymer chain relaxation.

4. Conclusion

The developed active film could be used to maintain the nutritional quality and extend shelf life of oils and related products, avoiding the addition of large amounts of food additives. Under the studied conditions, for each additive, a 2 wt.% concentration in the film was enough to provide the greatest antioxidant effect.

The *Quality Control* tool of the used software seems to be an interesting way to determine the induction time of degradation of edible oils. The 3009 cm^{-1} band, assigned to *cis* double bond, was the most sensitive in the determination of induction times using the pseudo-first order kinetics approach. In the set of bands corresponding to conjugated double bonds, only the *trans–trans* configuration band (987 cm^{-1}) was observed in the 60 °C condition even at short times in the control sample, indicating a high rate isomerization of *cis–trans* and *trans–cis* isomers to the *trans–trans* form.

BHT delays oil oxidation more efficiently than α-tocopherol, probably due to a synergic effect of the first one with the naturally occurring vitamin E in the soybean oil.

Results suggest that BHT releasing behavior depends on the hydrogen bonding interaction with the urethane group and it could be the aim of future works.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jfoodeng.2014.11.008.

References

Ahmed, M.K., Daun, J.K., Przybylski, R., 2005. FT-IR based methodology for quantitation of total tocopherols, tocotrienols and plastochromanol-8in vegetable oils. J. Food Compos. Anal. 18, 359–364.

Ahvenainen, R., 2003. Novel Food Packaging Techniques. Woodhead Publishing.

Anbinder, P.S., 2011. Caracterización, evaluación y aplicaciones de películas poliméricas activas. In: Facultad de Ciencias Exactas La Plata, Bs. As., Universidad Nacional de La Plata, Argentina.

Bart, J.C.J., 2005. Additives in Polymers: Industrial Analysis and Applications. Wiley and Sons Ltd.

Brody, A.L., Strupinsky, E.R., Kline, L.R., 2001. Active Packaging for Food Applications. CRC Press, Lancaster.

Chang, S.S., Senich, G.A., Smith, L.E., 1982. Migration of Low Molecular Weight Additives in Polyolefins and Copolymers: Final Report. National Bureau of Standards, NBSIR 82-2472.

Climaco-Pinto, R., Locquet, N., Eveleigh, L., Rutledge, D.N., 2010. Preliminary studies on the mid-infrared analysis of edible oils by direct heating on an ATR diamond crystal. Food Chem. 120, 1170–1177.

Destaillats, F., Angers, P., 2005. Thermally induced formation of conjugated isomers of linoleic acid. Eur. J. Lipid Sci. Technol. 107, 167–172.

EC, 2007. Commission Directive 2007/19/EC of 2 April 2007 amending Directive 2002/72/EC relating to plastic materials and articles intended to come into contact with food and Council Directive 85/572/EEC laying down the list of simulants to be used for testing migration of constituents of plastic materials and articles intended to come into contact with foodstuffs. L97, O.J.E.U. ed., pp. 50–69.

Frankel, E.N., 1993. In search of better methods to evaluate natural antioxidants and oxidative stability in food lipids. Trends Food Sci. Technol. 4, 220–225.

Frankel, E.N., 1998. Lipid Oxidation. The Oily Press, Dundee, UK.

Gómez-Alonso, S., Mancebo-Campos, V., Salvador, M.D., Fregapane, G., 2004. Oxidation kinetics in olive oil triacylglycerols under accelerated shelf-life testing (25–75 °C). Eur. J. Lipid Sci. Technol. 106, 369–375.

Gramiccioni, L., Prospero, P.D., Milana, M.R., Marzio, S.D., Marcello, I., 1986. Global migration from plastic materials into olive oil and isooctane: an experimental comparison. Food Chem. Toxicol. 24, 23–26.

Granda-Restrepo, D., Peralta, E., Troncoso-Rojas, R., Soto-Valdez, H., 2009. Release of antioxidants from co-extruded active packaging developed for whole milk powder. Int. Dairy J. 19, 481–488.

Guillén, M.D., Cabo, N., 1999. Usefulness of the frequency data of the fourier transform infrared spectra to evaluate the degree of oxidation of edible oils. J. Agric. Food Chem. 47, 709–719.

Guillén, M.D., Cabo, N., 2002. Fourier transform infrared spectra data versus peroxide and anisidine values to determine oxidative stability of edible oils. Food Chem. 77 (4), 503–510.

Innawong, B., Mallikarjunan, P., Irudayaraj, J., Marcy, J.E., 2004. The determination of frying oil quality using Fourier transform infrared attenuated total reflectance. Lebensm.-Wiss. u.-Technol. 37, 23–28.

Kadamne, J., Jain, V., Saleh, M., Proctor, A., 2009. Measurement of conjugated linoleic acid (CLA) in CLA-rich soy oil by attenuated total reflectance-fourier transform infrared spectroscopy (ATR-FTIR). J. Agric. Food Chem. 57, 10483–10488.

Koontz, J.L., Moffitt, R.D., Marcy, J.E., O'Keefe, S.F., Duncan, S.E., Long, T.E., 2010. Controlled release of α-tocopherol, quercetin, and their cyclodextrin inclusion complexes from linear low-density polyethylene (LLDPE) films into a coconut oil model food system. Food Addit. Contamin. 27, 1598–1607.

Lee, Y.S., Shin, H.S., Han, J.K., Lee, M., Giacin, J.R., 2004. Effectiveness of antioxidant-impregnated film in retarding lipid oxidation. J. Sci. Food Agric. 84, 993–1000.

Lerma-García, M.J., Ramis-Ramos, G., Herrero-Martínez, J.M., Simó-Alfonso, E.F., 2010. Authentication of extra virgin olive oils by Fourier-transform infrared spectroscopy. Food Chem. 118, 78–83.

Li, A., Yuan, B., Li, W., Wang, F., Ha, Y., 2013. Thermally induced isomerization of linoleic acid in soybean oil. Chem. Phys. Lipids 166, 55–60.

Maggio, R.M., Kaufman, T.S., Carlo, M.D., Cerretani, L., Bendini, A., Cichelli, A., Compagnone, D., 2009. Monitoring of fatty acid composition in virgin olive oil by Fourier transformed infrared spectroscopy coupled with partial least squares. Food Chem. 114, 1549–1554.

Marteau, C., Favier, D., Nardello-Rataj, V., Aubry, J.-M., 2014. Dramatic solvent effect on the synergy between α-tocopherol and BHT antioxidants. Food Chem. 160, 190–195.

Miltz, J., Hoojjat, P., Han, J.K., Giacin, J.R., Harte, B.R., Gray, I.J., 1988. Loss of antioxidants from high-density polyethylene. Its effect on oatmeal cereal oxidation. In: Hotchkiss, J.H. (Ed.), Food and Packaging Interactions. American Chemical Society, Washington, DC, pp. 83–93.

Moros, J., Roth, M., Garrigues, S., De la Guardia, M., 2009. Preliminary studies about thermal degradation of edible oils through attenuated total reflectance mid-infrared spectrometry. Food Chem. 114, 1529–1536.

Moya-Moreno, M.C.M., Mendoza-Olivares, D., Amézquita-López, F.J., Gimeno-Adelantado, J.V., Bosch-Reig, F., 1999. Analytical evaluation of polyunsaturated fatty acids degradation during thermal oxidation of edible oils by Fourier transform infrared spectroscopy. Talanta 50, 269–275.

Pardini, O.R., Amalvy, J.I., 2008. FTIR, ^{1}H NMR spectra and thermal characterization of water-based polyurethane/acrylic hybrids. J. Appl. Polym. Sci. 107, 1207–1214.

Peruzzo, P.J., Anbinder, P.S., Pardini, O.R., Costa, C.A., Leite, C.A., Galembeck, F., Amalvy, J.I., 2010. Polyurethane/acrylate hybrids: effects of acrylic content and thermal treatment on polymer properties. J. Appl. Polym. Sci. 116, 2694–2705.

Phipps, M., 1973. Liquid–liquid extraction of BHT from vegetable oils. J. Am. Oil Chem. Soc. 50, 21–23.

Pignitter, M., Stolze, K., Gartner, S., Dumhart, B., Stoll, C., Steiger, G., Kraemer, K., Somoza, V., 2014. Cold fluorescent light as major inducer of lipid oxidation in

soybean oil stored at household conditions for eight weeks. J. Agric. Food Chem. 62, 2297–2305.

Ritger, P., Peppas, N., 1987. A simple equation for description of solute release I. Fickian and non-fickian release from non-swellable devices in the form of slabs, spheres, cylinders or discs. J. Control. Release 5, 23–36.

Román-Falcó, I.P., Grané-Teruel, N., Prats-Moya, S., Martín-Carratalá, M.L., 2012. Kinetic study of olive oil degradation monitored by Fourier transform infrared spectrometry. Application to oil characterization. J. Agric. Food Chem. 60, 11800–11810.

Rooney, M.L., 2005. Introduction to active food packaging technologies. In: Han, J.H. (Ed.), Innovations in Food Packaging. Academic Press.

Schwope, A.D., Till, D.E., Ehntholt, D.J., Sidman, K.R., Whelan, R.H., Schwartz, P.S., Reid, R.C., 1987. Migration of BHT and Irganox 1010 from low-density polyethylene (LDPE) to foods and food-simulating liquids. Food Chem. Toxicol. 25 (4), 317–326.

Sherazi, S.T.H., Kandhro, A., Mahesar, S., Bhanger, M., Younis Talpur, M., Arain, S., 2009. Application of transmission FT-IR spectroscopy for the *trans* fat determination in the industrially processed edible oils. Food Chem. 114, 323–327.

Shlian, D.M., Goldstone, J., 1986. Toxicity of butylated hydroxytoluene. New England J. Med. 314, 648–649.

Van de Voort, F.R., 1992. Fourier transform infrared spectroscopy applied to food analysis. Food Res. Int. 25, 397–403.

Velasco, J., Dobarganes, C., 2002. Oxidative stability of virgin olive oil. Eur. J. Lipid Sci. Technol. 104, 661–676.

Vermeiren, L., Devlieghere, F., van Beest, M., de Kruijf, N., Debevere, J., 1999. Developments in the active packaging of foods. Trends Food Sci. Technol. 10, 77–86.

Vlachos, N., Skopelitis, Y., Psaroudaki, M., Konstantinidou, V., Chatzilazarou, A., Tegou, E., 2006. Applications of Fourier transform-infrared spectroscopy to edible oils. Anal. Chim. Acta 573–574, 459–465.

Wessling, C., Nielsen, T., Leufvén, A., Jägerstad, M., 1998. Mobility of α-tocopherol and BHT in LDPE in contact with fatty food simulants. Food Addit. Contamin. 15, 709–715.

Zhu, X., Lee, D.S., Yam, K.L., 2012. Release property and antioxidant effectiveness of tocopherol-incorporated LDPE/PP blend films. Food Addit. Contamin. 29 (3), 461–468.