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Chemistry and Physics of Lipids 134 (2005) 173–182
CPL
CHEMISTRY AND PHYSICS OF LIPIDS
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Direct monitoring of lipid oxidation in edible oils by Fourier transform Raman spectroscopy

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Received 2 February 2004; received in revised form 3 June 2004; accepted 13 January 2005

Abstract

Fourier transform Raman spectroscopy has been used to investigate the chemical changes taking place during lipid oxidation in several edible oils. Oxidative degradation of six vegetable oils was accelerated by heating at 160 °C. Formation of aldehydes was detected, and saturated as well as α,β-unsaturated aldehydes could be identified with the help of pure component spectra. The formation of conjugated double bond systems and the isomerisation of *cis* to *trans* double bonds was observed in the C=C stretching region and found to follow a distinct pattern for the different oils. It was possible to associate these differences to the fatty acid composition. The time-dependent intensity changes in certain Raman bands were compared to conventional parameters used to determine the extent of oxidation in oils, such as anisidine value and K_{270}, and showed good correlation.

Keywords: Lipid oxidation; Edible oils; Fourier transform Raman spectroscopy; *Cis–trans* double bonds; Conjugated dienes; α,β-Unsaturated aldehydes

1. Introduction

Due to health considerations there has been a significant shift in the sources of fats consumed in the last decades. Vegetable oils are increasingly important because of their high content in mono- and polyunsaturated fatty acids when compared to animal fats (Harwood et al., 1994). Oxidation of unsaturated fatty acids is the main reaction responsible of the degradation of lipids. Indeed, the oxidation level of oil and fat is an important quality criteria for food industry. Oxidation of oils not only produces rancid flavours but can also decrease the nutritional quality and safety by the formation of oxidation products, which may play a role in the development of diseases (Staprans et al., P1996a, b; Kanazawa et al., 2002). Under mild conditions, molecular oxygen reacts with the double bonds

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doi:10.1016/j.chemphyslip.2005.01.003

following a free radical mechanism, the so-called autoxidation. This process is generally well understood, but is quite complex and variable in edible oils due to its dependency on oil type and conditions of oxidation. Thermal stress speeds up oxidative reactions, and thus, lipid oxidation is a major concern in frying operations.

Many methods have been developed to access the extent of oxidative deterioration, which are related to the measurement of the concentration of primary or secondary oxidation products or of both. The most commonly used are peroxide value (PV) that measures volumetrically the concentration of hydroperoxides, anisidine value (AV), spectrophotometric measurement in the UV region and gas chromatographic (GC) analysis for volatile compounds (Frankel, 1998a). Since hydroperoxides are primary oxidation products, the method is only really applicable for samples that are oxidized to low levels and under mild conditions. The AV method aims to monitor secondary oxidation products by reaction with *p*-anisidine and spectrophotometric measurement at 350 nm. This test measures high-molecular weight saturated and unsaturated carbonyl compounds, principally 2-enals. The specific absorptions at 232 nm (K_{232}) and at 270 nm (K_{270}) are related to the content of conjugated dienes and trienes, respectively. The above-mentioned methods have been established over time and have found wide spread applications as routine methods to determine oxidative deterioration of lipids. Common properties of this traditional analytical methods are that they provide a single read-out (index), which despite of being useful do not provide information on the actual chemical composition of the sample. Contrary to these methods, more advance and time consuming analytical techniques like gas chromatography have been used to study the oxidation process in more detail, mostly focusing on volatile compounds (Snyder et al., 1988). Recently, also the determination of epoxy compounds in thermoxidized oils has been reported (Velasco et al., 2002). These methods are sensitive to detect and characterize low levels of oxidation in various oil and food lipids; however, they require substantial sampling handling and in general are time consuming.

Vibrational spectroscopy, because of its high content in molecular structure information, has also been considered to be useful for the fast measurement of lipid oxidation. In contrast to time consuming chromatographic methods, modern techniques of IR and Raman spectrometry are rapid and do not require any sample preparation steps prior to analysis. Fourier transform (FT) mid-infrared (IR) spectroscopy gives information about the different functional groups present in the sample; thus, it is not limited to just one kind of compound like the indices mentioned above. This technique has been used to monitor oil oxidation under moderate and accelerated conditions and the major band changes have been interpreted (van de Voort et al., 1994). Numerous papers followed this work, which all aimed to quantify certain parameters that are related to oil oxidation with IR spectroscopy, including peroxide value (Guillén and Cabo, 2002; Ruiz Medina et al., 2001), anisidine value (Dubois et al., 1996), volatile compounds (Ahro et al., 2002) and malondialdehyde (Mirghani et al., 2002). Some of these parameters have also been determined by near-infrared spectroscopy (Yildiz et al., 2001).

Unlike infrared, Raman spectroscopy is poorly explored in its application to edible oils. It has been basically applied to characterization (Baeten et al., 1998) and authentication (Marigheto et al., 1998); and to the quantitative analysis of total unsaturation, *cis*/*trans* isomers (Sadeghi-Jorabchi et al., 1991) and free fatty acid content (Muik et al., 2003). But to our best knowledge, there is no work published describing the spectral changes during oil oxidation. Raman spectroscopy, like IR spectroscopy, probes molecular vibrations, but the measurement principle is totally different. Since the selection rules applying for Raman spectroscopy are opposed to those applying for infrared, other vibrational modes can be observed. Qualitatively, asymmetric vibrational modes and vibrations of polar groups are more likely to exhibit prominent IR absorption, while symmetric vibrational modes generally give rise to strong Raman scattering. Therefore, Raman and infrared are considered complementary techniques. When oxidation of oils is monitored using infrared spectroscopy the presence of *trans* double bonds as well as the formation of polar compounds such as hydroperoxides and carbonylic compounds can be clearly detected. Using Raman spectroscopy chemical changes affecting C=C bonds and the carbon skeleton of the lipids during oxidation are expected to be reflected in spectral changes.

In this work, the changes in the Raman spectra of six edible vegetable oils during oxidation were measured to explore if the oxidation process follows the

same pattern in the different types of oil under study. The samples studied included oils with different proportions of oleic, linoleic and linolenic acyl groups. The aim of this study was to monitor the oxidation process in the different samples and to characterize the spectral changes occurring as lipid oxidation proceeds. Thus, it highlights the capability of Raman spectroscopy to provide information about the chemical processes taking place during lipid oxidation in a rapid and experimentally simple way.

2. Experimental

2.1. Instrumentation

Raman spectra were recorded with a Bruker RFS (FT)-Raman spectrometer fitted with a liquid nitrogen cooled Ge detector. The samples were illuminated by a Nd:YAG laser line at 1064 nm with a power of 500 mW using a focussed laser beam. All spectra were recorded with a resolution of 4 cm^{-1} and averaged over 200 scans. Acquisition of one spectrum took 5 min.

2.2. Samples

The six vegetable oils, virgin olive, corn, sunflower, rapeseed, peanut and safflower, were purchased from Austrian and Spanish supermarkets.

2.3. Oxidation and Raman measurements

About 200 ml of the oils were heated at 160 (±2) °C and stirred continuously on a magnetic stirrer for 9 h. Every half an hour 4 ml were sampled, left to cool to room temperature for recording the FT-Raman spectra. No sample preparation was required. The rest of the sample was deep-frozen for the reference measurements.

2.4. Reference measurements

The fatty acid composition of the oils was measured with the gas chromatographic method according to the IUPAC method 2302 with preparation of the fatty acid methyl esters according to IUPAC method 2301 (“special method”) (Paquot and Hautfenne, 1987). Iodine and peroxide values were determined according to the IUPAC methods 2205 and 2501, respectively (Paquot and Hautfenne, 1987). To measure the spectrophotometric characteristics (K_{232} and K_{270}) weighed oil samples were diluted in cyclohexane.

2.5. Reagents

Cyclohexane, chloroform, carbon tetrachloride, acetic acid, Hanus-reagent (0.2N) and sodium thiosulfate were provided by Panreac (Barcelona, Spain). Isooctane, potassium iodide, *p*-anisidine, hexanal (Capronaldehyde), *trans*-2-hexenal and *trans*, *trans*-2,4-decadienal were purchased from Fluka (Buchs, Switzerland). *Trans*-9,10-epoxystearic acid and *cis*-9,10-epoxystearic acid were provided by Sigma-Aldrich (Steinheim, Germany).

3. Results

The Raman spectra of lipids contain predominantly bands arising from vibrations of the hydrocarbon chains, with some contributions from those of polar groups. Fig. 1 shows the Raman spectra of virgin olive, rapeseed and safflower oils. The major bands in the spectral region below 1800 cm^{-1} can be found at about 1267 cm^{-1} (symmetric rock in *cis* double bond δ(=C–H)), 1302 cm^{-1} (in-phase methylene twist), 1442 cm^{-1} (scissoring mode of methylene $\delta(CH_2)$), 1655 cm^{-1} (*cis* double bond stretching ν(C=C)) and 1747 cm^{-1} (ester stretching ν(C=O)). The region between 2850 and 2980 cm^{-1} is characteristic of the symmetric and asymmetric C–H stretching vibrations of methyl and methylene groups. At about 3007 cm^{-1} the symmetric, =C–H stretching vibration can be found. The major differences among the spectra of the different oils are located at the bands 1267, 1655 and 3007 cm^{-1}, which correspond to vibrations in *cis* double bonds. The intensities of these bands reflect the total unsaturation and correlate with the iodine values of the oils. Table 1 shows the fatty acid composition of the six vegetable oils used in this study. They differ in the percentage of saturated, mono- and polyunsaturated fatty acids, the iodine values ranging between 80 for virgin olive and 166 for safflower oils.

3.1. Changes in Raman spectra during oxidation

Fig. 2 shows the regions of the most prominent changes in the Raman spectrum of sunflower oil during

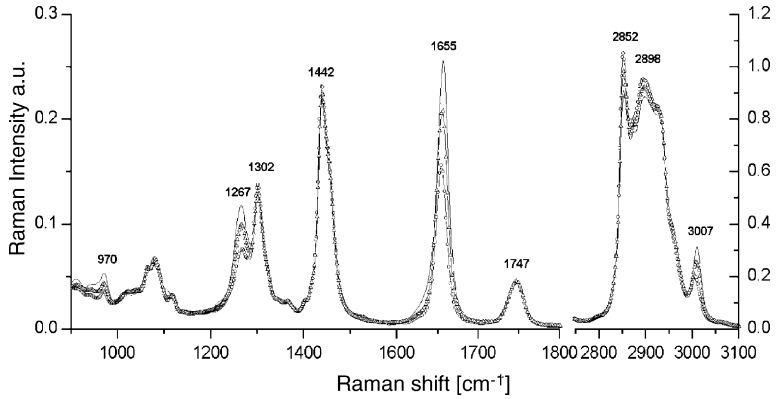

Fig. 1. Raman spectra of (—) safflower, (△) rapeseed and (◯) virgin olive oil.

the oxidation process. Generally, the bands which were not involved in C=C losses or isomerisations, did not change during the oxidation process. Small changes compared to the intensity in the region between 2830 and 2990 cm^{-1} were observed for all the oils, but the interpretation of these changes is very difficult, due to the origin of these bands. In none of the oils a change of the C=O ester band was observed, which indicates that hydrolysis is not taking place. Hydrolysis occurs at higher temperatures (>180 °C) and in presence of moisture. It was therefore decided to use the intensity of this band to normalize the intensities of the bands shown in figures where band evolutions are presented (I_n/I_{1745}). This was done in order to better visualize the general tendency of the bands to increase, decrease or to be shifted. The normalization corrected for small intensity variations across the whole spectrum that occurred due to changes of the density and viscosity of the oil.

The spectrum of virgin olive oil showed initially a band located at 1525 cm^{-1} which can be assigned to the C=C stretching in highly conjugated systems, like carotenoids. These compounds are present in virgin olive oil but not in the refined oils. Carotenoids have antioxidant properties as radical scavengers and singlet oxygen quenchers in lipid oxidation (Yanishlieva et al., 1998). The antioxidative behaviour of carotenoids is closely related to its own oxidation and the loss of carotenoids could be clearly observed at the beginning of the heating period in the band at 1525 cm^{-1}, which disappeared after 90 min.

3.1.1. Formation of secondary oxidation products

At temperatures above 100 °C, the primary oxidation products, the hydroperoxides exist only transiently and decompose rapidly into a multitude of volatile and non-volatile products. Alcohols, saturated aldehydes, α,β-unsaturated aldehydes and epoxy compounds have been reported as major secondary oxidation products (Frankel et al., 1992). The Raman spectra of three aldehydes and two epoxy compounds were measured in order to gain insight in their characteristic spectral features. These molecules were thus used as reference compounds for secondary oxidation products.

Table 1
Fatty acid compositions and iodine values of the six vegetable oils used in this study

	Olive	Peanut	Rapeseed	Corn	Sunflower	Safflower
C 16:0 + C 18:0	12.7	11.3	6.0	12.0	10.2	6.6
C 18:1	81.2	50.2	64.4	31.9	30.4	19.5
C 18:2	4.8	33.2	19.5	54.7	59.1	70.7
C 18:3	0.6	2.1	9.8	1.0	0.2	0.9
IV	80	99	110	128	123	166

C 16:0, palmitic acid; C 18:0, stearic acid; C 18:1, oleic acid; C 18:2, linoleic acid; C 18:3, linolenic acid. Values for acids reflect percentage of sum; IV, iodine value.

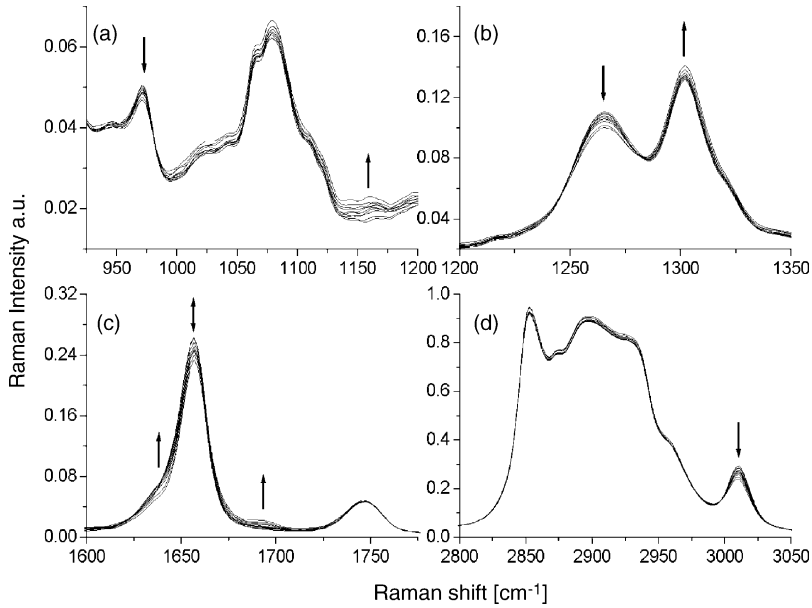

Fig. 2. Regions of the most prominent changes in the Raman spectrum of sunflower oil during 9 h oxidation: (a) 900–1200 cm^{-1}, (b) 1200–1350 cm^{-1}, (c) 1600–1775 cm^{-1} and (d) 2800–3050 cm^{-1}.

Trans-9,10- and *cis*-9,10-epoxystearic acids are oxidation products of the oleic acid with the oxirane group at the double bond site. Apart from the C–H stretching region both compounds exhibit bands of medium intensity at 1064, 1295 and 1443 cm^{-1}. Unfortunately, they coincide with the major bands of the olive oil and other bands present in the spectra are very weak. Consequently, the identification of this kind of compounds in the oxidized oils was not possible.

Hexanal exhibits a weak band at 1724 cm^{-1}, which corresponds to the C=O stretching of the aldehyde. In *trans*-2-hexenal, a secondary oxidation product from linolenate decomposition, the C=O stretching band of the aldehyde is much more intense and shifted to a lower wave number (1687 cm^{-1}). This shift and also the gain in intensity are due to conjugation with the C=C bonds (Lin-Vien et al., 1991). The Raman intensity of the C=O stretching vibration in this α,β-unsaturated aldehyde is even stronger than the very strong C=C stretching band located at 1639 cm^{-1}. *Trans*, *trans*-2,4-decadienal, a major decomposition product from heated oxidized linoleate, exhibits a very strong band at 1641 cm^{-1} which corresponds to the C=C stretching vibration in conjugated systems and a strong band at 1164 cm^{-1}, which is assigned to the accompanying C–C stretching vibration. Furthermore, the spectrum has a strong band at 1684 cm^{-1}, which corresponds to the C=O stretching of the aldehyde.

For all the studied oils, a broad band appears in the region between 1680 and 1730 cm^{-1} as oxidation proceeds, as can be seen in Fig. 2c for sunflower oil. To better visualize the changes, the spectrum of the non-oxidised oil ($t = 0$) was subtracted from all the others. In this way, one broad band at around 1690 becomes clearly visible (see Fig. 3). Furthermore, in the region around 1725 cm^{-1} a slight increase can be noticed, which can be better discerned in rapeseed oil (Fig. 3b). With the help of the spectra of the reference compounds, these bands can be assigned. At 1725 cm^{-1}, the C=O stretching of saturated and around 1690 cm^{-1} the C=O stretching of conjugated unsaturated aldehydes.

3.1.2. General loss in cis double bonds

The most characteristic changes in the lipid structure during oxidation are the loss of unsaturation due to attack of oxygen and the following radical reactions. The general loss in *cis* double bonds during the oxidation process can be observed in the bands 3007 and 1267 cm^{-1} in all the oils. These bands remain unaltered

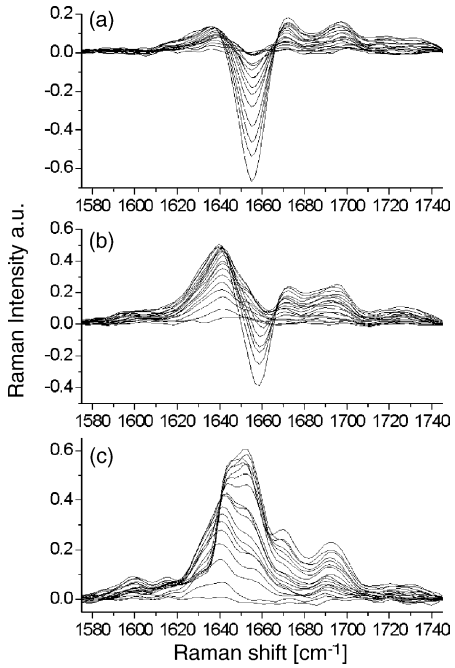

Fig. 3. Changes in the C=C and C=O stretching region during the oxidation process for: (a) olive oil, (b) rapeseed oil, (c) sunflower oil. Spectra have been normalized to the intensity at 1745 cm^{-1} and the spectra of the non-oxidized oils ($t = 0$) have been subtracted.

at the beginning of the heating period. In olive oil, the duration of this stability period (90 min) can be correlated directly to the carotenoids peak. In the other oils, this period lasts longest for corn (120 min) and shortest for sunflower oil (60 min) and is most probably due to added antioxidants like tocopherols. The ensuing decrease in the bands is more pronounced in rapeseed and peanut oil probably due to the higher content in linolenic acid, which is much more prone to oxidation than linoleic and oleic acid (Frankel, 1998b). Apart from the above-mentioned bands, a decrease in the band at 970 cm^{-1} is observed. This band was assigned to *trans* double bonds in a spectroscopic study of fatty acid methyl esters in the autoxidation of alkyd resin coatings (Agbenyega et al., 1991). Nevertheless, *trans* double bonds are formed during the oxidation process, and therefore, it should increase. So we conclude that the band at 970 cm^{-1} corresponds more probably to the out of phase HC=CH wag vibration of *cis* double bonds, which must also be expected at this frequency. Fig. 2 shows the changes in all these bands for sunflower oil. Concerning the *cis* double bond stretching ν(C=C), band at 1655 cm^{-1}, the decrease is only clearly visible in olive oil. For the rest of the oils this band changes in shape and increases in intensity (see Fig. 3). These changes and the apparent increase in the degree of unsaturation are due to different ν(C=C) bands, arising from *trans* and conjugated double bonds generated during the oxidation process, that overlap with the band due to *cis* double bonds.

3.1.3. Formation of trans and conjugated double bonds

Thermal oxidation of unsaturated oils is accompanied by considerable isomerisation of double bonds leading to products containing *trans* double bonds and conjugated double bond systems (Frankel, 1998b). In Fig. 3, changes in the C=C stretching region during the oxidation process for olive, rapeseed and sunflower oil are shown, as examples. For a better visualization, the spectra of the non-oxidized oils ($t = 0$) have been subtracted. In all oils, the formation of a band at 1670 cm^{-1} can be seen. This band has been previously described and assigned to isolated *trans* olefins (Sadeghi-Jorabchi et al., 1991). Another band increases at 1640 cm^{-1}, which is assigned to conjugated dienes. In Fig. 4, the evolution of these bands in the different oils are presented. In oil samples with low content in polyunsaturated acids less conjugated dienes are formed, this is especially noticeable in olive oil where only a slight increase of the band at 1640 cm^{-1} can be observed. Furthermore, small differences in linolenic acid content lead to strong increases of the band at 1640 cm^{-1}, as can be seen in rapeseed oil and also in the comparison of corn and sunflower oil, which have very similar contents in oleic and linoleic acid. Sunflower oil has lower linolenic content and this is reflected in a lower increment of this band.

The changes in the band around 1655 cm^{-1} are the most difficult to interpret. In olive oil, this band is stable at the beginning and decreases after 4 h of heating. In rapeseed and peanut oil, a slight increase is followed by a decrease after 6 h. In sunflower, corn and safflower oil, the band is steadily increasing and only at the end of the observation time a slight decrease is visible (see Fig. 4c). The behaviour of the band in olive oil can be attributed to the decrease in *cis* double bonds. But to explain the increase of the band in the rest of the oils, the formation of a compound which gives rise to an intense

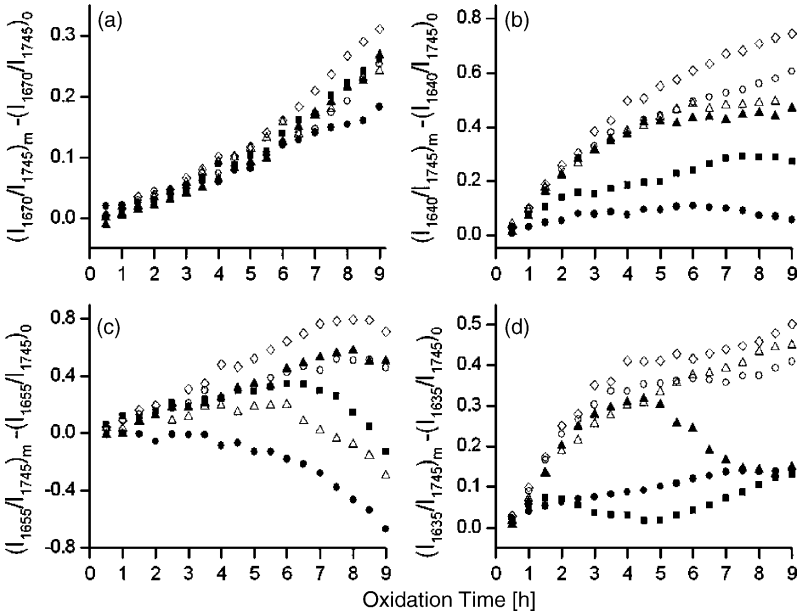

Fig. 4. Raman band evolution during 9 h oxidation at: (a) 1670 cm^{-1}, (b) 1640 cm^{-1}, (c) 1655 cm^{-1}, (d) 1635 cm^{-1} of (●) olive, (■) peanut, (△) rapeseed, (◯) corn, (▲) sunflower and (◊) safflower oil.

band at a close frequency must be considered. Looking at the difference spectra, this band can be located at about 1653 cm^{-1}. Due to its position and intensity, the most probable explanation is that it derives from conjugated dienes. This is plausible because conjugated dienes formed in the oxidation process of oils can occur in different isomeric configurations, which appear at different frequencies in the Raman spectra and give strong Raman scattering. Furthermore, according to the different behaviour of the oils this band is related to the ratio oleic/linoleic acid. The lower the ratio the higher the increase at this frequency. Other than the band at 1640 cm^{-1}, this band seems to be independent of the linolenic acid content.

Another band in this region appears, which can be easily identified in the subtracted spectra of sunflower oil. It is found at 1635 cm^{-1} and due to its position at lower frequency than the others it can be tentatively assigned to the C=C stretching vibration of conjugated trienes. Again the behaviour of this band is different in the studied oils (see Fig. 4d). Olive and peanut oil do not show a significant increase of this band in the whole heating period. Safflower, rapeseed, corn and sunflower oil show a rapid increase until 4–5 h of heating. Then they show a moderate increase except sunflower oil, which exhibits a strong diminution. This different behaviour cannot solely be explained with the fatty acid composition of the oils.

Concerning spectral differences due to the appearance of conjugated double bond systems, another spectral region is worth to be mentioned. It is situated between 1135 and 1180 cm^{-1}, where the C–C stretching vibration in conjugated systems is found. This band remains unaltered in olive oil and increases for the rest, safflower oil exhibiting the highest increase. It seems that also this band consists in at least two different vibrations, but they are difficult to separate because the band and the changes are of low intensity.

3.2. Correlation between spectral changes and conventional parameters to determine the extent of oxidation

To evaluate the oxidation state of the oil, the following parameters have been measured in selected samples. The spectrophotometric absorption at 232 and 270 nm, expressed as K_{232} and K_{270}, measures the formation of conjugated dienes and trienes, respectively. The iodine value is a measure of the total number of double bonds and the anisidine value measures high-

molecular weight saturated and unsaturated carbonyl compounds in triacylglycerols.

During the oxidation process, a general decrease in iodine value is observed for all the oils. This decrease correlates very well with the behaviour of the spectral bands at 3007 and 1267 cm^{-1} mentioned above.

The anisidine value increases for all oils during the heating. For olive oil this increase is much lower than for the other oils, which is not surprising as the respond of this test not only depends on the amount of aldehydic components but also on their structure. It has been found that a double bond in the carbon chain conjugated with the carbonyl double bond increases the molar absorbance four to five times. This means that 2-alkenals and 2,4-dialkenals will contribute more to the anisidine value than saturated and other unsaturated aldehydes. Oxidative degradation of oleic acid gives less α,β-unsaturated aldehydes than that of linoleic and linolenic acid, which explains the lower response of olive oil. A high correlation ($R^2 = 0.96$) was found between the anisidine value and the intensity of the band at 1693 cm^{-1} in the Raman spectra (see Fig. 5a), which was valid for all the oils independent of their composition. Apart from this correlation, the Raman spectra also contain information about the content in saturated aldehydes at the band at 1725 cm^{-1}, which increases for all oils during the heating period, but is not correlated to the anisidine value.

The spectrophotometric absorption at 270 nm showed for all the oils a tendency that resembles very well that of the band located at 1635 cm^{-1}. Furthermore, good correlation ($R^2 = 0.92$) was found between K_{270} and the intensity at 1635 cm^{-1} (see Fig. 5b), which confirmed the assignment of this band to conjugated trienes. The K_{232} value increased strongly in all the oils with high percentage of polyunsaturated fatty acids as oxidation proceeded. For olive oil this parameter increased very slightly, reflecting that few conjugated dienes are formed. No correlation was found between this parameter and any of the Raman bands in the C=C stretching region changing during oxidation. This is mainly due to the strong decrease in the *cis* double bonds, but also confirms the fact that conjugated dienes give rise to more than one Raman band in this region due to the existence of different isomers. Nevertheless, correlation can be found with the C–C stretching region at 1157 cm^{-1} ($R^2 = 0.86$) (see Fig. 5c).

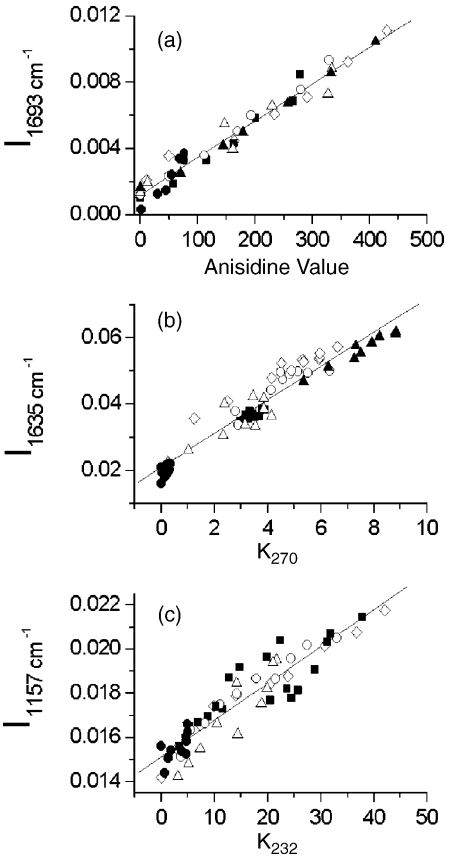

Fig. 5. Correlation between the intensity of selected Raman bands and reference measurements to determine the extent of oxidation: (a) anisidin value vs. intensity at 1693 cm^{-1}; (b) K_{270} vs. intensity at 1635 cm^{-1}; (c) K_{232} vs. intensity at 1157 cm^{-1}. (●) Olive, (■) peanut, (△) rapeseed, (◯) corn, (▲) sunflower and (◊) safflower oil.

In the conditions used for the oxidation study hydroperoxides are only formed transitorily. This was reflected in the peroxide value, which did not reach elevated values in none of the oils. Consequently, there was no correlation found between peroxide values and frequencies in the Raman spectra.

3.3. Information obtainable by Raman spectroscopy

The information available in the Raman spectra can be successfully correlated to conventional parameters used to determine the rate of oxidation in thermally stressed oils. The anisidin value and the *K* values can be estimated simultaneously from Raman spectra of the

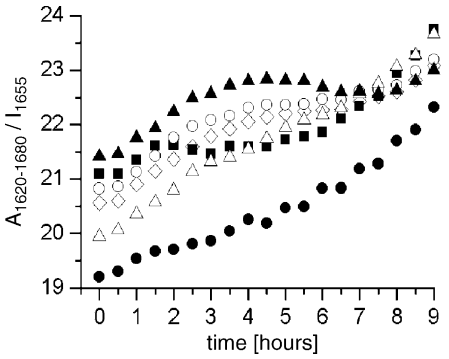

Fig. 6. Changes of the ratio $A_{1620–1680}/I_{1655}$ with the oxidation time. (●) Olive, (■) peanut, (△) rapeseed, (◯) corn, (▲) sunflower and (◊) safflower oil.

oils. In addition, the degree of unsaturation can also be determined using the Raman spectra. The determination of this parameter using Raman spectroscopy has already been reported using univariate and multivariate calibration. It should be mentioned that the approach of Sadeghi-Jorabchi et al. (1990), which uses the ratio of intensities between the band at 1660 and 1444 cm^{-1} cannot be employed in thermally stressed oils due to the changes that happen in the C=C stretching region during oxidation.

According to the results obtained in this study, Raman spectroscopy has been found to be useful for monitoring oil oxidation, as it allows insight in the different behaviour of the investigated oils under oxidative conditions. The major differences between the oils were found in the band at 1655 cm^{-1} which corresponds to the C=C stretching region. This band comprises the formation of conjugated double bond systems, of *trans* double bonds and the loss of *cis* double bonds. Thus, it reflects a major part of conformational and configurational changes, which the oils undergo during the oxidation process. This information can be summarized using the ratio of the area between 1620 and 1680 cm^{-1} and the height of the band at 1655 cm^{-1} ($A_{1620–1680}/I_{1655}$), both calculated with respect to a baseline point located at 1800 cm^{-1}. The evolution of this parameter is shown in Fig. 6 for all the oils. For safflower, peanut, corn, rapeseed and sunflower oil, the ratio $A_{1620–1680}/I_{1655}$ increases initially, and then reaches a plateau and after a period, which depends on the nature of the oil, increases again. This complex behaviour reflects the formation of compounds containing conjugated dienes and trienes, and isomerisations that take place during thermal oxidation. For virgin olive oil, the initial value of this parameter is much lower than for the other oils. The initial higher value of this parameter can be attributed to small amounts of conjugated dienes and trienes produced by deodorization at elevated temperatures in refined oils, which is also reflected in K_{270} and K_{232} of these oils. Furthermore, in olive oil, the ratio $A_{1620–1680}/I_{1655}$ behaves differently; it increases steadily because few conjugated compounds are formed. Thus, for olive oil this parameter principally reflects the isomerisation of *cis* double bonds to *trans* double bonds.

This study demonstrates the utility of Raman spectroscopy in following the oxidative degradation of lipids in edible oils. The high molecular specific information content in the Raman spectra allows to gain insight in the nature of chemical changes taking place during oxidation. Again it shall be stressed that practically no sample preparation is required in Raman spectroscopy. Furthermore, using micro-sampling techniques like Raman microspectroscopy also spatially resolved measurements are feasible. In further work, we plan to apply this technique to the study of related oxidation processes in biological systems that are gaining increased attention with regard to possible connection between lipid oxidation and pathological events.

Acknowledgement

B.M. thanks the Spanish Ministerio de Asuntos Exteriores for a Ph.D. fellowship.

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