

Detection of olive oil adulteration by corn oil addition applying ATR-FTIR spectroscopy

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Abstract

The main objective of this study was to assess the potential of Attenuated Total Reflectance - Fourier Transform Infrared (ATR-FTIR) spectroscopy for detection of olive oil (OO) adulteration by corn oil (CO) addition. The differences recorded for the band around 3006 cm^{-1} assigned to the C-H stretching vibration of the cis-double bonds (=CH) can be used for quantification of OO adulteration. The height of the 3006 cm^{-1} band for pure OO sample is obviously smaller than for pure CO or for its mixtures with OO. The maximum absorbance value of the band at this frequency changed according to the extent of OO adulteration. Also, the position of this band has shifted versus the percentage of CO added in OO from 3009 to 3005 cm^{-1} . The maximum heights of the two bands at ~ 3006 and 2922 cm^{-1} were also used in assessing of OO adulteration. The 2922 cm^{-1} band is assigned to the symmetric stretching vibration of the aliphatic $-\text{CH}_2$ groups. The calculated ratio of the two peak heights express the percentage of the hydrogen-carbon bonds linked by cis-double bonds (=CH) present in the oil samples. There was recorded a high correlation ($R^2=0.91$) between the ratio of these peak heights and the percentage of CO added to OO. Based on calibration curve absorbance versus the percentage of adulterant in OO it is possible to assess the OO samples' adulteration. Significant positive correlations ($R^2>0.94$) were found at some specific frequencies i.e. 3011, 3006 and 722 cm^{-1} , respectively, while for 1096 and 912 cm^{-1} it was recorded R^2 values of 0.9. The combined frequency regions 3020–3000, 1100–900, 750–700 could be successfully used for quantification the OO counterfeit by CO addition as a possible adulterant agent. Based on our results it can be noticed that ATR-FTIR spectroscopy could be successfully used as a rapid, non-destructive technique with a minimum sample preparation for detection of OO adulteration.

Keywords: olive oil, corn oil, adulteration, ATR-FTIR spectroscopy

1. Introduction

OO contains a series of compounds that are very beneficial to most functions of the human body and its' biological and therapeutic value is related in many aspects to its chemical structure. OO is considered as one of the valuable edible oils due to its capacity to prevent and reduce the incidence of cancer and cardiovascular diseases [1, 2]. Olive oils are one of the oils that have been subjected to adulteration for a long time.

The OO adulteration with different cheaper vegetable oils (i.e. sunflower oil, soybean oil, corn oil) is a danger both for economic reasons as well as for OO consumers' health. Although the adulteration is done for economic reason, the action affects the quality of the olive oil [3]. From this point of view, OO authenticity has become a serious issue for producers, consumers and also for authorities [4].

Addition of CO in OO resulted in significant changes in the content of polyunsaturated (PUFAs), monounsaturated (MUFAs) and saturated fatty acids SAFAs. Thus, is expected to appear various spectral changes that could be detected using ATR-FTIR spectroscopy. The application of FTIR spectroscopy has increased in food studies, and particularly has become a powerful analytical tool in the study of edible oils and fats [5]. By applying FTIR can be possible the qualitative determination of organic compounds based on the bands appeared in the infrared spectrum at a specific frequency caused by the characteristic vibration of each molecular group [6-8]. Nowadays, it is evident the special focus towards evaluation the potential of FTIR spectroscopy in order to assess the OO adulteration based on the spectral changes at specific wavenumbers [9-11]. There have been performed several studies concerning the characterization, classification, and authentication of edible fats and oils using infrared spectroscopy [12, 13]. The study carried out by Vlachos *et al.* [6], indicates that the limit of detection for olive oil adulteration is 9% if the adulterant is corn oil or sesame seed oil while it is lower (6%) if the adulterant agent is sunflower oil or soybean oil. These results emphasize the usefulness of FTIR spectroscopy in detecting and quantifying of the spectral changes as well as the advantages in terms of speed and expense per analysis. The power of FTIR as a quantitative tool consists in its ability to readily carry out the multi-component analyses [14-16]. Taking into account these issues, the goal of this study was to explore the potential of FTIR spectroscopy for detection of OO adulteration by CO addition.

Abbreviations

ATR-FTIR spectroscopy: Attenuated Total Reflectance - Fourier Transform Infrared spectroscopy; OO: olive oil; CO: corn oil; SAFAs: saturated fatty acids; MUFAs: mono unsaturated fatty acids; PUFAs: poly unsaturated fatty acids.

2. Materials and Methods

Oil samples. In this study were used extra virgin olive oil (OO) and corn oil (CO) as well as OO adulterated samples by deliberately adding of CO in the range 10-100% (v/v) from 10 to 10%. These mixtures were manually shaken for homogenization. OO and CO were purchased from local hypermarket (Timisoara, Romania).

OO was produced in Italy by PIETRO CORICELLI SPA, Spoleto and CO was produced in Romania by SC MAN SRL, Satu Mare.

FTIR analysis. A FTIR-8400S Fourier Transform Infrared Spectrophotometer (*Shimadzu*, Japan) equipped with attenuated total reflectance (ATR) accessory was used to record the FTIR spectra for pure OO and CO as well as for their mixtures in different proportions. Oil samples were sequentially measured at room temperature (25°C) in the range 400–4000 cm^{-1} with scanning time 60 s and 2 cm^{-1} resolution. These spectra were obtained against the background of air spectrum. After every scan of oil sample, a background of new reference air spectrum was performed. Also, the ATR plate was cleaned with a soft tissue soaked in acetone for any residues removing from previous samples. The ATR cleanliness was assessed by comparing the obtained background spectrum to the previous one. All spectra were processed with the computer software program Spectrum for Windows XP Professional (Shimadzu).

Statistical analysis. Simple linear regression analysis was applied using the Origin 7.0 software program for obtaining of some correlations between absorbance at specific frequencies and the percentage of CO added in OO.

3. Results and discussion

The recorded FTIR spectra of both pure OO and CO samples as well as their mixtures in different percentages (80%OO+20%CO; 60%OO+40%CO; 40%OO+60%CO and 20%OO+80%CO) are shown in Figure 1. Also, the FT-IR spectra of pure oil samples (OO, CO) and their mixture (50%OO+50%CO) are presented in Figure 2. In Figure 3 are shown the spectral bands at specific frequencies obtained for investigated pure and adulterated oils.

The FTIR spectra recorded for oil samples present specific bands with different intensities and forms. Based on the differences in the intensity and the frequency at which the maximum absorbance of the bands appears, closely related to the nature and composition of the investigated oil samples, we will try to assess the OO adulteration by CO addition. From Figures 1 and 2 can be noticed that there are notable differences in the absorption intensity obtained for pure OO and CO and their mixtures. We have noticed that in the spectral region 4000-3100 cm^{-1} the oil samples showed no infrared absorption.

In the spectral interval 3100–2800 cm^{-1} it can be noticed that both pure and adulterated oil samples showed specific bands.

In agreement with the research conducted by Poiana *et al.*, 2012 [17], in this region occur bands close to the frequency 2965, 2935, 2895 and 2855 cm^{-1} assigned to the symmetrical and asymmetrical C–H stretching vibration of the CH_2 and CH_3 aliphatic groups due to the alkyl rest of triglycerides that exist in large quantities in vegetable oils. Besides these information, the first think we can notice about these analyzed spectra is the difference of the bands around 3006 cm^{-1} assigned to the C–H stretching vibration of the *cis*-double bond ($=\text{CH}$). The oil composition, which is different for OO, CO or their mixtures, affects the exact position of the bands and also affects the occurred shifts when the proportion of the fatty acid is changed. Data from Table 1 highlight the shift of the band position versus the percentage of CO added in OO. Thus, the band position significantly varied from 3009 to 3005 cm^{-1} with the extent of OO adulteration by CO addition. For example, as show in Table 1, pure OO sample has a maximum at 3005 cm^{-1} while by adding of CO the maximum shifts to higher frequencies.

In order to make possible the quantifying of OO adulteration, the maximum heights of the two bands at ~ 3006 and 2922 cm^{-1} were also used. The 2922 cm^{-1} band is assigned to the symmetric stretching vibration of the aliphatic $-\text{CH}_2$ groups. The calculated ratio of the two peak heights express the percentage of the hydrogen–carbon bonds linked by *cis*-double bonds ($=\text{CH}$) present in the oil samples. Is interesting to notice that there is a high correlation ($R^2=0.91$, Figure 4) between the ratio of the peak heights and the CO percentage added to OO. For pure OO (Figures 1 and 2) we can seen that the absorbance at 3011 cm^{-1} , specific frequency of the methyl-linoleate, is lower (0.1944) than the value recorded at 3006 cm^{-1} , specific to the methyloleate (0.2221). Our results outline that by increasing of CO percentage added in OO it was induced an increase in absorbance value recorded at 3011 cm^{-1} due to increasing of linoleic acid content in triglycerides composition. Moreover, the height of the band at 3006 cm^{-1} for pure OO sample is obviously smaller than those values registered for pure CO or for its mixtures with OO. When the percentage of CO added in OO

increased, the height of this band also increased, as is shown in Table 1. The height of the band that appears at 2922 cm^{-1} suffers small changes compared to the 3006 cm^{-1} band.

The results obtained here are consistent with those reported by Vlahos *et al.* [6] that emphasized that the absorption close to 3010 cm^{-1} can be useful for assessment of OO adulteration by adding of other foreign oils.

In the interval 1800–1600 cm^{-1} appear two important bands close to 1745 and 1660 cm^{-1} . The absorbance at 1743 cm^{-1} is a significant characteristic to the oils with high short carbohydrate chain and saturated fatty acids content. The absorbance value at this wavenumber does not show significant variations by varying the percentage of CO added in OO because of the low content of saturated acids in both OO and CO.

The second band from this spectral range occurs at 1654 cm^{-1} due to the double $-\text{C}=\text{C}-$ bond and can be correlated with the content of polyunsaturated fatty acids from the oil samples [18]. Based on recorded data it doesn't possible to quantify the addition of CO in pure OO because the signal intensity at this frequency is not show a uniform variation to the percentage of CO added in OO.

In the range 1600–1390 cm^{-1} occurs only a single spectral band close to 1440–1445 cm^{-1} assigned to the vibrations of deformation $\delta(\text{C}-\text{H})$. This band can be used for total unsaturation assessing. For FT-IR spectra of pure OO sample it was recorded a maximum at 1464 cm^{-1} (0.5934) and for pure CO at 1466 cm^{-1} (0.5941). Thus, there were not notable differences in the signal registered at this frequency with the percentage of CO added to OO.

Table 1. The shift of the band around 3006 cm^{-1} versus the CO (%) added to OO

Percentage of adulterant (%)	Wavenumber (cm^{-1})	Absorbance
0	3005.1950	0.2222
10	3005.1950	0.2342
20	3006.1594	0.2332
30	3006.1595	0.2347
40	3006.1595	0.2423
50	3006.1595	0.2324
60	3007.1239	0.2497
70	3007.1239	0.2596
80	3008.0884	0.2697
90	3009.0528	0.2994
100	3009.0528	0.3050

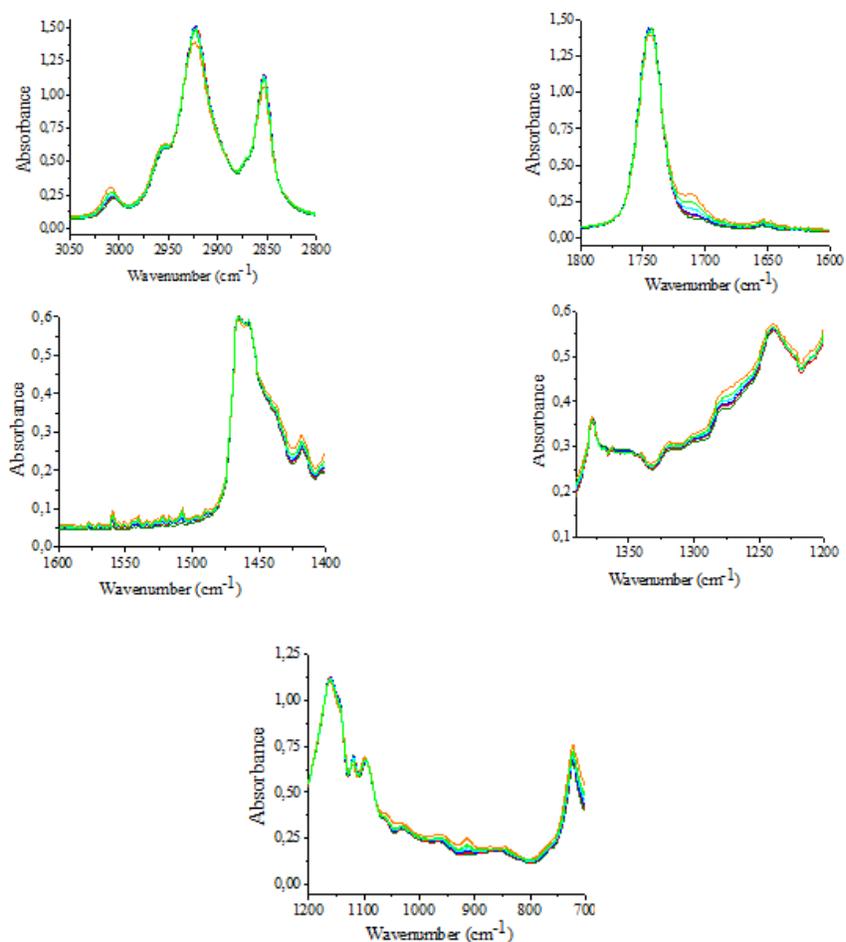
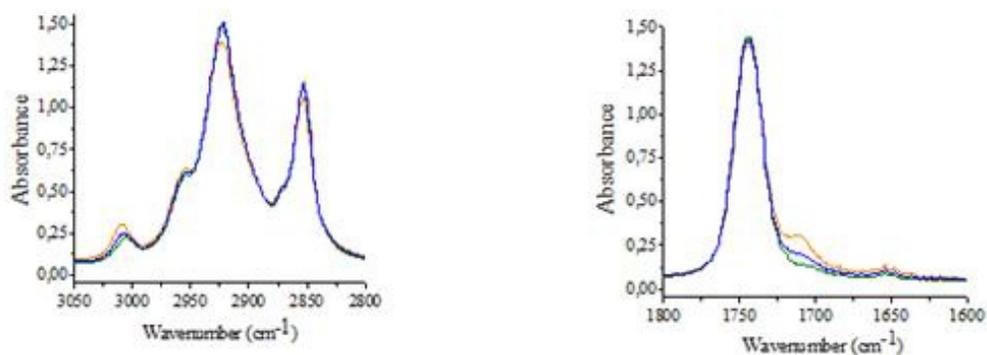


Figure 1. Comparativ FT-IR spectra regions of pure OO, CO and their mixtures: pure OO (—); 80%OO+20%CO (—); 60%OO+40%CO (—); 40%OO+60%CO (—); 20%OO+80%CO (—); pure CO (—).



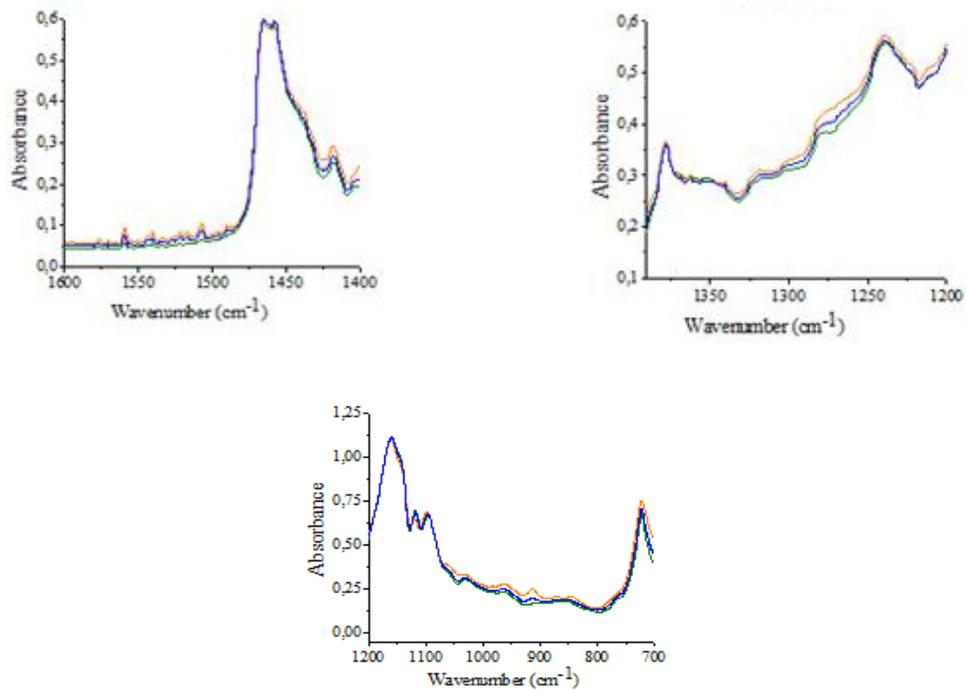


Figure 2. Comparative FT-IR spectra regions of pure OO, CO and mixture: pure OO (—); pure CO (—); 50%OO+50%CO (—).

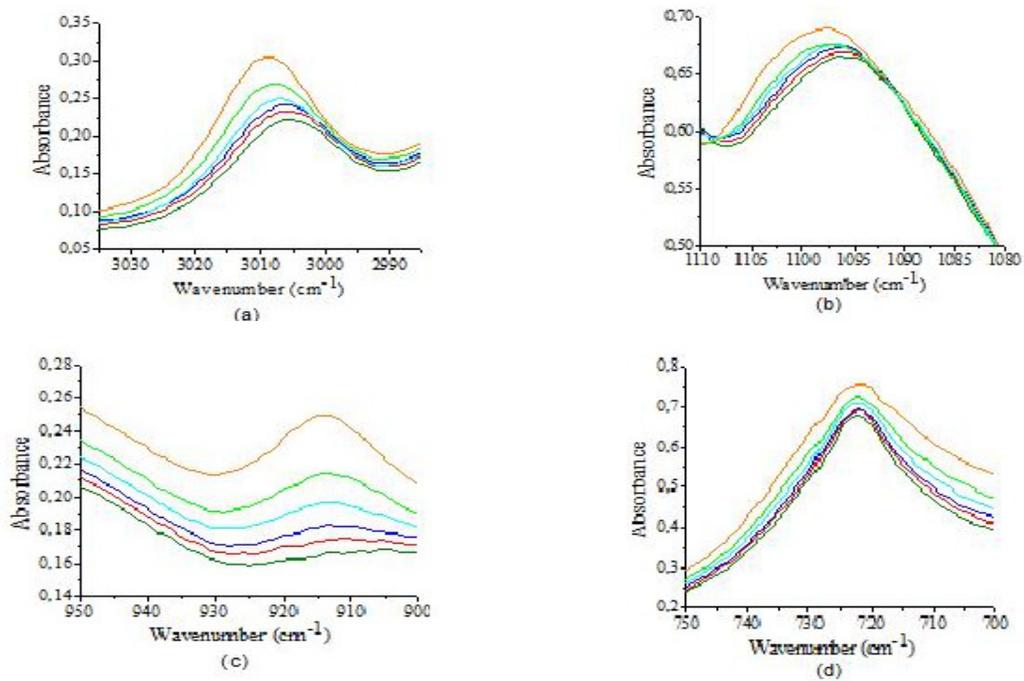


Figure 3. FT-IR spectra regions of pure OO, CO and their mixtures: pure OO (—); 80%OO+20%CO (—); 60%OO+40%CO (—); 40%OO+60%CO (—); 20%OO+80%CO (—); pure CO (—).
(a: A_{3006} ; b: A_{1096} ; c: A_{912} ; d: A_{722})

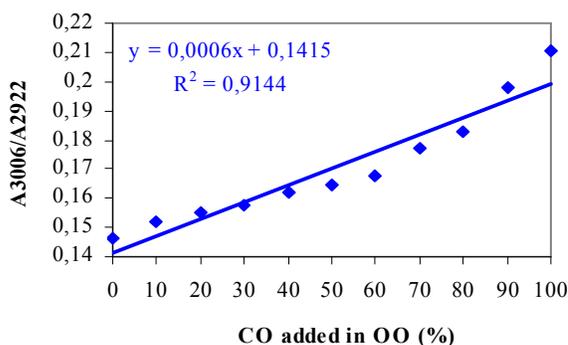


Figure 4. Monitoring the OO adulteration with CO using the ratio A3006/A2922

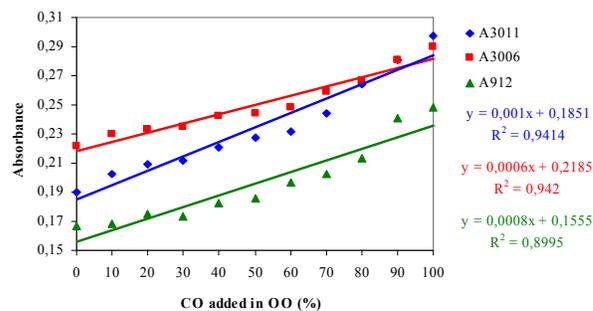
In the region $1390\text{--}1200\text{ cm}^{-1}$ it can be noticed two bands: the first one, close to frequency of 1303 cm^{-1} , was assigned to deformation vibration of methylene group ($-\text{CH}_2-$) and the second one at 1270 cm^{-1} is specific to deformation vibration in the plan of the group $=\text{CH}-$ from the unconjugated *cis* double bonds.

From the obtained FTIR spectra it was possible to identify two maximums: at wavenumber of 1378 cm^{-1} and 1238 cm^{-1} , respectively. The intensity of the absorption at 1378 cm^{-1} is similar for both oils (OO and CO), thus, the absorbance recorded at this frequency is not useful in detection of OO adulteration. These results are in line with those reported by Alexa *et al.* [18]. The absorbance recorded at 1238 cm^{-1} for pure CO was slowly higher than the value registered for pure OO sample, but there were not significant differences in these values with the increasing of percentage of adulterant.

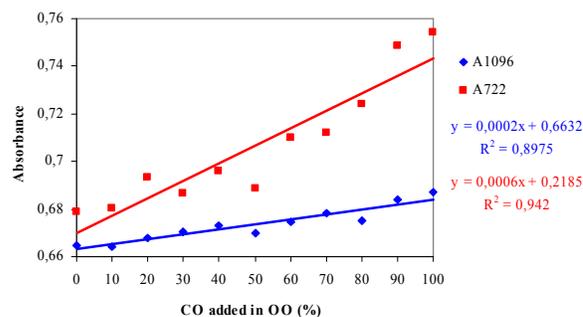
In the spectral range $1200\text{--}700\text{ cm}^{-1}$ were noticed bands in the ranges $1100\text{--}1000\text{ cm}^{-1}$ and $900\text{--}800\text{ cm}^{-1}$ assigned to vibration of C–C bonds of carbohydrate chain from oil composition. Also, other bands occur at $1150\text{--}1060$ and $970\text{--}800\text{ cm}^{-1}$ and these are specific to C=O bonds' vibration. Also, in Figure 5 are shown the calibration curves absorbance at specific frequencies (3011, 3006, 1096, 912 and 722 cm^{-1}) versus percentage of CO added in pure OO.

These results emphasize that some specific frequencies could be used to appreciate the extent of OO adulteration by CO addition. Significant positive correlations ($R^2 > 0.94$) was found at frequencies of 3011, 3006 and 722 cm^{-1} (Figure 5). Also, at wavenumbers of 1096 and 912 cm^{-1} the correlation coefficient values were $R^2 = 0.9$. This means that the combined frequency regions 3020--

3000 , $1100\text{--}900$, $750\text{--}700$ could be successfully used for quantification the OO forgery by CO addition. This remark is in agreement to the results of study performed by Rohman & Man [19].



(a)



(b)

Figure 5. Linear regression absorbance versus percent of CO added in OO

(a: A_{3011} , A_{3006} and A_{912} ; b: A_{1096} and A_{722})

4. Conclusions

ATR-FTIR spectroscopy is a rapid, non-destructive technique with minimum sample preparation that offers an easy way to assess the extent of OO adulteration by CO addition, as cheaper adulterant. The absorbance recorded at different specific frequencies could be successfully used for assessing the OO forgery by CO addition but only after a proper calibration. The oil samples composition affects the exact position of the bands and also affects the occurred shifts. The differences of the band around 3006 cm^{-1} assigned to the C-H stretching vibration of the *cis*-double bonds ($=\text{CH}$) can be used for quantification of OO adulteration. The absorbance of OO samples varied at this frequency with the increasing of adulteration degree. Also, the position of the band recorded at this frequency has shifted versus the percentage of CO added to OO. Thus, the maximum of this band significantly varied from 3009 to 3005 cm^{-1} with the extent of OO adulteration. The maximum heights of

the two bands at ~ 3006 and 2922 cm^{-1} were also used for assessment of OO adulteration.

The calculated ratio of the two peak heights express the percentage of the hydrogen-carbon bonds linked by cis-double bonds (=CH) present in the oil sample. Our data outline that there is a high correlation ($R^2=0.91$) between the ratio of the peak heights and the percentage of CO added to OO. The combined frequency regions 3020–3000, 1100–900, 750–700 could be successfully used for quantification the OO counterfeit by CO addition, as a possible adulterant agent.

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Compliance with Ethics Requirements

Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human and/or animal subjects (if exists) respect the specific regulations and standards.

References

1. Frankel, E.N., Nutritional and biological properties of extra virgin olive oil. *Journal of Agricultural and Food Chemistry* **2011**, *59*(3), 785–92.
2. Kontogianni, M.D.; Panagiotakos, D.B.; Chrysohoou, C.; Pitsavos, C.; Zampelas, A.; Stefanadis, C., The impact of olive oil consumption pattern on the risk of acute coronary syndromes: The Cardio 2000 case-control study, *Clinical Cardiology* **2007**, *30*(3), 125–129.
3. Christy, A.A.; Kasemsumran, S.; Du, Y.; Ozaki, Y., The detection and quantification of adulteration in olive oil by Near-Infrared Spectroscopy and Chemometrics, *Analytical Sciences* **2004**, *20*(6), 935–940.
4. Ben-Ayed R., Kamoun-Grati N., Rebai A. An Overview of the Authentication of Olive Tree and Oil, *Comprehensive reviews in Food Science and Food Safety* **2013**, *12*(2), 218–227.
5. Guillen, M.D., & Cabo, N., Some of the most significant changes in the Fourier transform infrared spectra of edible oils under oxidative conditions, *Journal of the Science of Food and Agriculture* **2000**, *80*(14), 2028–2036.

6. Vlachos, N.; Skopelitis, Y.; Psaroudaki, M.; Konstantinidou, V.; Chatzilazarou, A.; Tegou, E., Applications of Fourier transform-infrared spectroscopy to edible oils, *Analytica Chimica Acta* **2006**, *573–574*, 459–465.
7. Allam M, Hamed SF: Application of FT-IR spectroscopy in the assessment of olive oil adulteration, *Journal of Applied Sciences Research* **2007**, *3*(2), 102–108.
8. Gurdeniz, G.; Tokatli, F.; Ozen, B., Differentiation of mixtures of monovarietal olive oils with mid-infrared spectroscopy and chemometrics, *European Journal of Lipid Science and Technoogy* **2007**, *109*(12), 1194–1202.
9. Obeidat, S.M.; Khanfar, M.S.; Obeidat, W.M., Classification of edible oils and uncovering adulteration of virgin olive oil using FTIR with the aid of chemometrics, *Australian Journal of Basic and Applied Sciences* **2009**, *3*(3), 2048–2053.
10. Allam, M.; Hamed, S.F., Application of FT-IR spectroscopy in the assessment of olive oil adulteration, *Journal of Applied Sciences Research* **2007**, *3*(2):102–108.
11. Lerma-Garcia, M.J.; Ramis-Ramos, G.; Herrero Martínez, J.M.; Simo-Alfonso, E.F., Authentication of extra virgin olive oils by Fourier-transform infrared spectroscopy, *Food Chemistry* **2010**, *118*(1), 78–83.
12. Rohman, A.; Che Man, Y.B., Fourier transform infrared (FTIR) spectroscopy for analysis of extra virgin olive oil adulterated with palm oil, *Food Research International* **2010**, *43*(3), 886–892.
13. Zhang, Q.; Liu, C.; Sun, Z.; Hu, X.; Shen, Q.; Wu, J., Authentication of edible vegetable oils adulterated with used frying oil by Fourier Transform Infrared Spectroscopy, *Food Chemistry* **2012**, *132*(1), 1607–1613.
14. Safar, M.; Bertrand, D.; Robert, P.; Devaux, M.F.; Genot, C., Characterization of edible oils, butters and margarines by Fourier transform infrared spectroscopy with attenuated total reflectance, *Journal of the American Oil Chemists' Society* **1994**, *71*(4), 371–377.
15. Rodriguez-Saona, L.E.; Allendorf, M.E., Use of FTIR for rapid authentication and detection of adulteration of food, *Annual Review of Food Science and Technology* **2011**, *2*, 17.1–17.17.
16. Lai, Y.W.; Kemsley, E.K.; Wilson, R.H., Potential of Fourier Transform Infrared Spectroscopy for the authentication of vegetable oils, *Journal of Agricultural and Food Chemistry* **1994**, *42*(5), 1154–1159.
17. Poiana, M.A.; Mousdis, G.; Alexa E.; Moigradean, D.; Negrea, M.; Mateescu, C., Application of FT-IR spectroscopy to assess the olive oil adulteration, *Journal of Agroalimentary Processes and Technologies* **2012**, *18*(4), 277–282.
18. Alexa, E.; Dragomirescu, A.; Pop, G.; Jianu, C.; Dragos, D., The use of FT-IR spectroscopy in the identification of vegetable oils adulteration, *Journal of Food, Agriculture & Environment* **2009**, *7*(2), 20–24.
19. Rohman, A.; Che, Man Y.B., Quantification and classification of corn and sunflower oils as adulterants in olive oil using chemometrics and FTIR spectra, *The Scientific World Journal* **2012**, Article ID 250795, 6 pages, [doi:10.1100/2012/250795](https://doi.org/10.1100/2012/250795).