Comparative Study of Thermal Stability of Olive Oil and Sesame Oil Using FTIR Spectroscopy and UV-Vis Spectroscopy

S.I.M. Ifthikar [1#], R.W.U.D. De Silva [1], R. Shanthini [1]

[1]*Department of Chemical and Process Engineering, University of Peradeniya, Sri Lanka*

1. Introduction

Frying involves immersing food in oil at 170–220°C, a popular method at homes, restaurants (food services), street food vending and at large industrial operations for its convenience and unique effects on food flavor and texture. Prolonged high-temperature exposure causes oil to undergo cis-trans isomerization, thermo-oxidation, polymerization, and hydrolysis, forming over 400 heat-induced reaction products, most of which are absorbed by the food (Paul & Mittal, 1996; Poiana et al., 2015; Sunisa W. et al., 2011). Heating supplies the activation energy required for oxidation, forming compounds like aldehydes, alcohols, esters, short-chain hydrocarbons, and volatile ketones that can significantly affect the flavor, aroma, and nutritional value of the oils (Gonçalves et al., 2014). However, Antioxidants can counteract oxidation by protecting target lipids from initiators and stalling the propagation phase of lipid oxidation (Laguerre et al., 2007).

Olive oil contains antioxidants like tocopherols, chlorophylls, carotenoids, lutein, and phenolic compound (Frankel, 2010), while sesame oil contains sesamin, sesamolin, chlorophylls and carotenoids (Borchani et al., 2010; Gandul-Rojas & Minguez-Mosquera, 1996; Wan et al., 2015). Oils with high antioxidant content exhibit greater oxidative stability; however, high-temperature treatments can deplete these antioxidants, reducing the oil's stability (Cai et al., 2021).

According to studies (Li et al., 2013), cis to tarns isomerization occurs when the heating temperature of oil is increased to 150°C since trans double bonds are thermodynamically more stable than cis double bonds (Brühl, 2014). FFAs are formed from triacylglycerol by ester bond cleavage due to lipase action, high temperature, and moisture. They act as pro-oxidants by accelerating hydroperoxide breakdown, leading to further oxidation and undesirable flavors in oils. FFA levels are used to assess oil quality and edibility (Chew & Nyam, 2019).

Although many studies have investigated the thermal stability of coconut oil (Omar et al., 2014; Sharanke & Sivakanthan, 2022; Yu et al., 2018), there has been limited research on the thermal stability of olive oil during prolonged frying and to our knowledge no study on the thermal stability of sesame oil during prolonged frying.

A study by Abenoza et al., (2016), analyzed the changes in various physicochemical and nutritional parameters of two different olive oils (Picual and Arbequina) during short-term frying at 180°C for 3 minutes. Another study compared the thermal stability of different varieties of olive oil during long-term frying over 27 hours, evaluating free acidity, peroxide and p-anisidine values, specific extinction coefficients, oxidative stability, fatty acids, vitamin E, β-carotene, and total phenols in oil (Casal et al., 2010).

Most of these studies are directed towards analysis by high-performance liquid chromatography (HPLC), a method that requires toxic solvents or high volumes of solvents for quantification, which is more expensive and unhealthy for the laboratory staff and the community.

In contrast, our study employs FTIR and UV-visible spectroscopy, widely used methods for qualitative analysis due to their simplicity, speed, and eco-friendliness (Rohman & Che Man, 2013; Tótoli & Salgado, 2015). Known for its minimal use of solvents and harmful chemical reagents, FTIR and UV visible spectroscopy is a staple in "green analytical chemistry". To our knowledge, no studies in the literature compare the thermal stability of olive oil and sesame oil. Therefore, this study aims to validate an environmentally friendly spectrophotometric method for analyzing the thermal stability of oils by examining spectral changes in functional groups resulting from thermal degradation and to compare the thermal stability of olive oil and sesame oil subjected to prolonged frying (185 ± 3°C for 4, 8, and 16 hours).

2. Materials and Methods

2.1 Samples

Extra virgin olive oil, extra virgin coconut oil, and sesame oil were purchased from the local market in Kandy, Sri Lanka. Coconut oil (CO) and sesame oil (SO) were packaged in transparent polyethylene terephthalate (PET) bottles, whereas olive oil (SO) was packaged in a colored glass bottle. According to the label the olive oil was obtained directly from olives and solely by mechanical means, and coconut oil was cold-pressed. Tests for CO were done considering it as a dummy sample to validate the use of FTIR spectroscopy, for the comparison of the thermal stability of oils.

2.2 Heat treatment of oil samples

Oil samples (25.0 ± 0.1 g) were weighed in Pyrex Petri dishes (9 cm diameter) and heated at 185 ± 3°C for 4, 8, and 16 hours in the dark using an electric oven (Memmert, UN160) with temperature control. After heating, samples were cooled to room temperature and stored in the dark to prevent photodegradation until analysis. Heat treatments were performed in duplicates.

2.3 FTIR spectra acquisition

All spectra of duplicate oil samples were scanned using FTIR spectrometers (Perkin Elmer, FTIR spectrometer, spectrum two). The instrument was connected to the spectrum IR Software. 32 scans at a resolution of 1 cm^{-1}, were used to capture FTIR spectra in the 4000-400 cm^{-1}, region. These spectra were subtracted against the background of the air spectrum.

2.4 UV-Vis spectra acquisition

OO and SO samples were scanned using UV-visible spectroscopy (Agilent Cary 8454) at the region of 720-380 nm (visible region) at the resolution of 1 nm and a scan rate of 6000.00 (nm/min). Before each scan, the equipment was zeroed considering air as the reference sample (Alves et al., 2019).Approximately 3/4 of the cuvette (volume-3500 μl, Optical path length- 10 mm, Outer dimensions(L×H×W)-12.5mm×45mm×12.5mm) was filled with oil sample (without dilution) and scanned at room temperature (26 °C). After each scan, absorption peaks were recorded for each oil sample.

2.5 Statistical Analysis

Design of experiments (DOE) via factorial designs was a technique in Minitab (Product version: Minitab® 19.1 (64-bit)) that was used to determine the influence of individual and interactive effects on the output results.

Table 1. Factor information of the DOE factorial design used

Factor	Levels	Values
Oil type	3	CO
		SO
		OO
Heating time	3	0h
		4h
		8h

The software called The Unscrambler® X (version 10.4) and Origin was used to do PCA (Principle Component Analysis), a statistical approach for downscaling multidimensional data to smaller dimensions while preserving the majority of the original data (Karamizadeh et al., 2013).

2.6 Fatty acid analysis

To assess the oxidative stability of OO, CO, and SO with heating time, the acid value of fresh and heated samples was determined using a laboratory test (da Silva Oliveira et al., 2019; Nielsen, 2017)

3. Results and Discussions

3.1 FTIR Spectroscopy Analysis

Fig. 1, 2, and 3 show the FTIR spectra for the samples OO, SO, and CO respectively. The FTIR spectrum for SO heated for 16 hours was not obtained due to its transformation into a bright yellow gel-like consistency, making it unsuitable for FTIR analysis, which is better suited for liquid samples. Analysis was done on the absorption bands at six different wavenumbers that are connected to oil oxidation and the cis- and trans-isomerization of double bonds. These wave numbers are 3006 cm^{-1} (assigned to C–H stretching symmetric vibration of the cis double bonds, =CH), 1743 cm^{-1} (due to the formation of secondary oxidation products and the formation of epoxides), 987 cm^{-1} (due to formation of conjugated trans isomer), 967 cm^{-1} (assigned to out-of-plane bending vibration of trans –HC=CH– group of di-substituted olefins), 885 cm^{-1} (due thermal decomposition of methyl oleate hydroperoxides and are attributed to trans epoxides) and 722 cm^{-1} (assigned

to overlapping of CH2 rocking vibration and the out-of-plane vibration of cis –HC=CH– group of di-substituted olefins) (Poiana et al., 2015b).
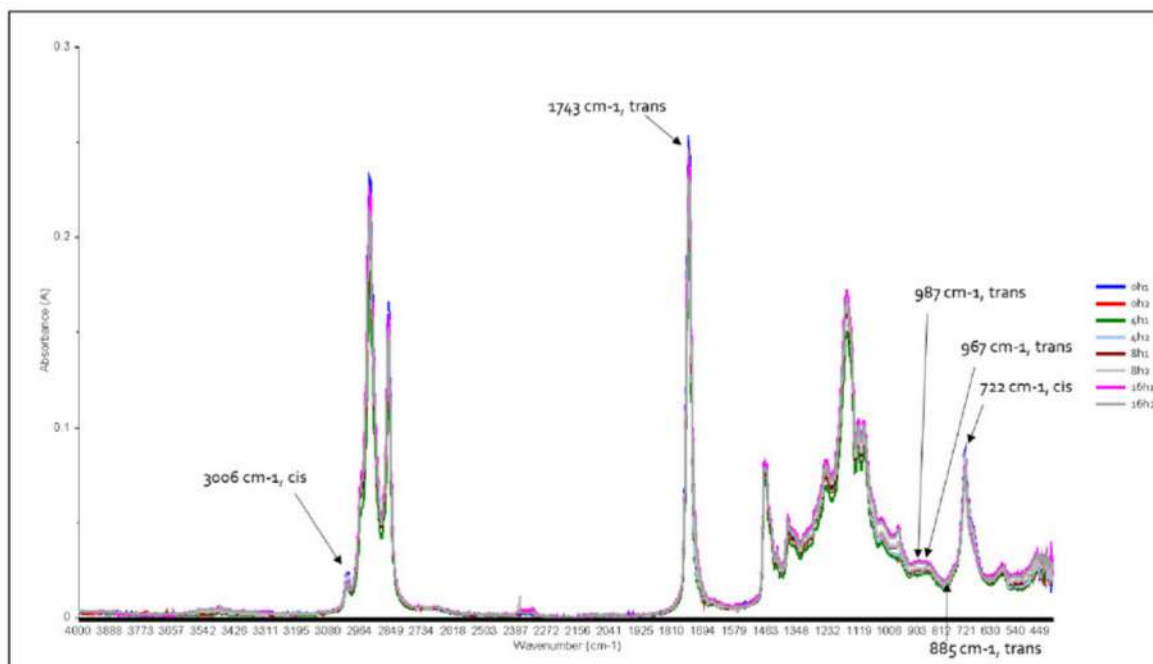

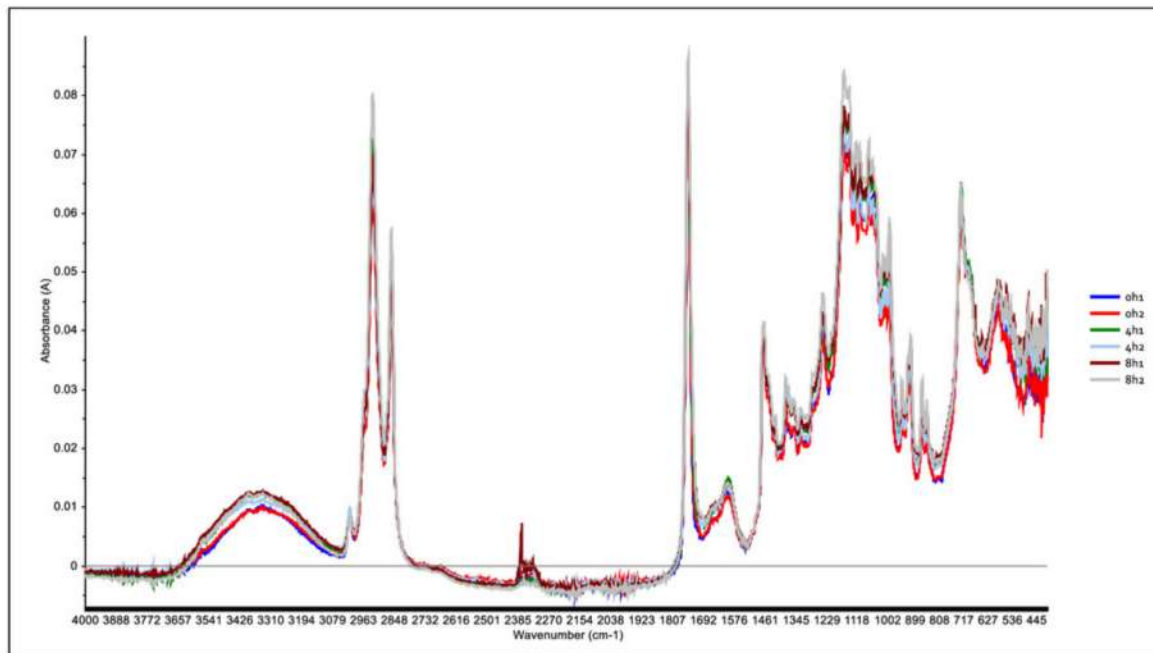**Fig. 1.** FTIR spectrum of OO at room temperature

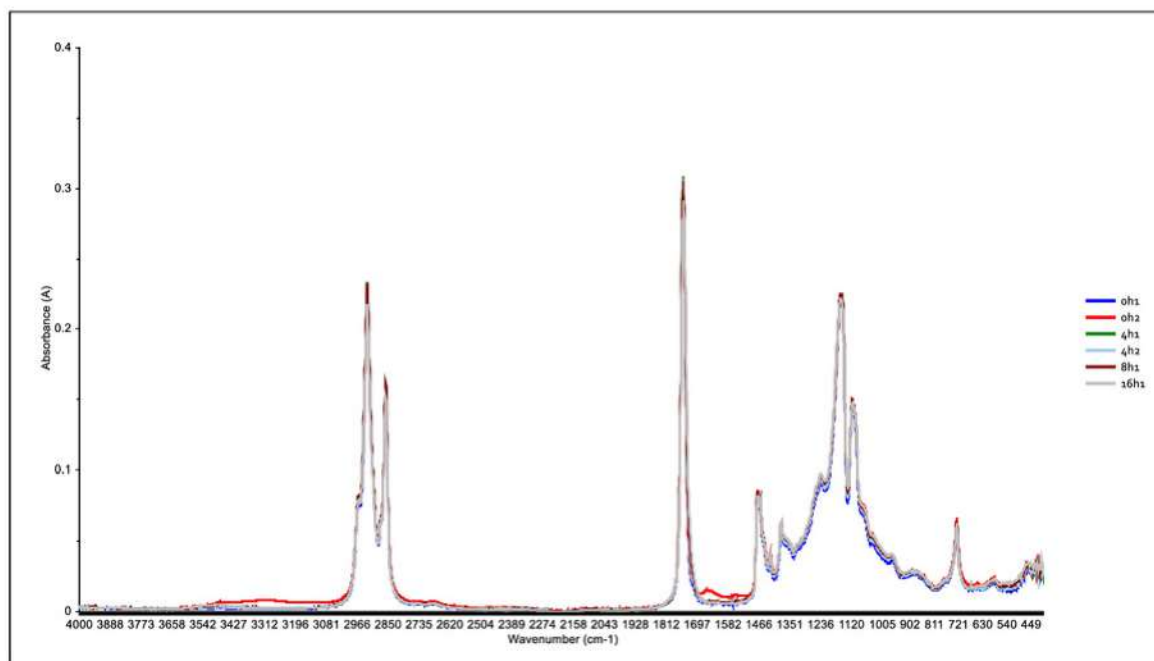**Fig. 2.** FTIR spectrum of SO at room temperature

Fig. 3. FTIR spectrum of CO at room temperature

While the absorbance at wavenumbers 3006 and 722 cm^{-1} belongs to the cis double bonds of unsaturated fatty acids and has been utilized for their evaluation (Guille & Cabo, 2002; Poiana et al., 2015b), the absorbance at wavenumbers 1743, 967, 987, and 885 cm^{-1} has been widely employed to determine the level of trans unsaturated fatty acid in oils. Fig. 4. (a) and (e) demonstrate a progressive decrease in absorbance with increasing heat treatment at 3006 cm^{-1} and 722 cm^{-1}, respectively for OO. This indicates the disappearance of double bonds in the cis conformation. On the other hand, Fig. 4. (c) shows the absorption bands recorded at 967 and 987 cm^{-1} in response to the formation of trans isomers (non-conjugated trans and conjugated trans, respectively) in SO. The loss of the cis double bonds, due to their breakdown to produce secondary oxidation products as a result of heat-induced oil oxidation can be observed in Fig. 4. (b). Consequently, heat stress has caused cis-trans isomerization of double bonds and oil oxidation in both SO and OO.
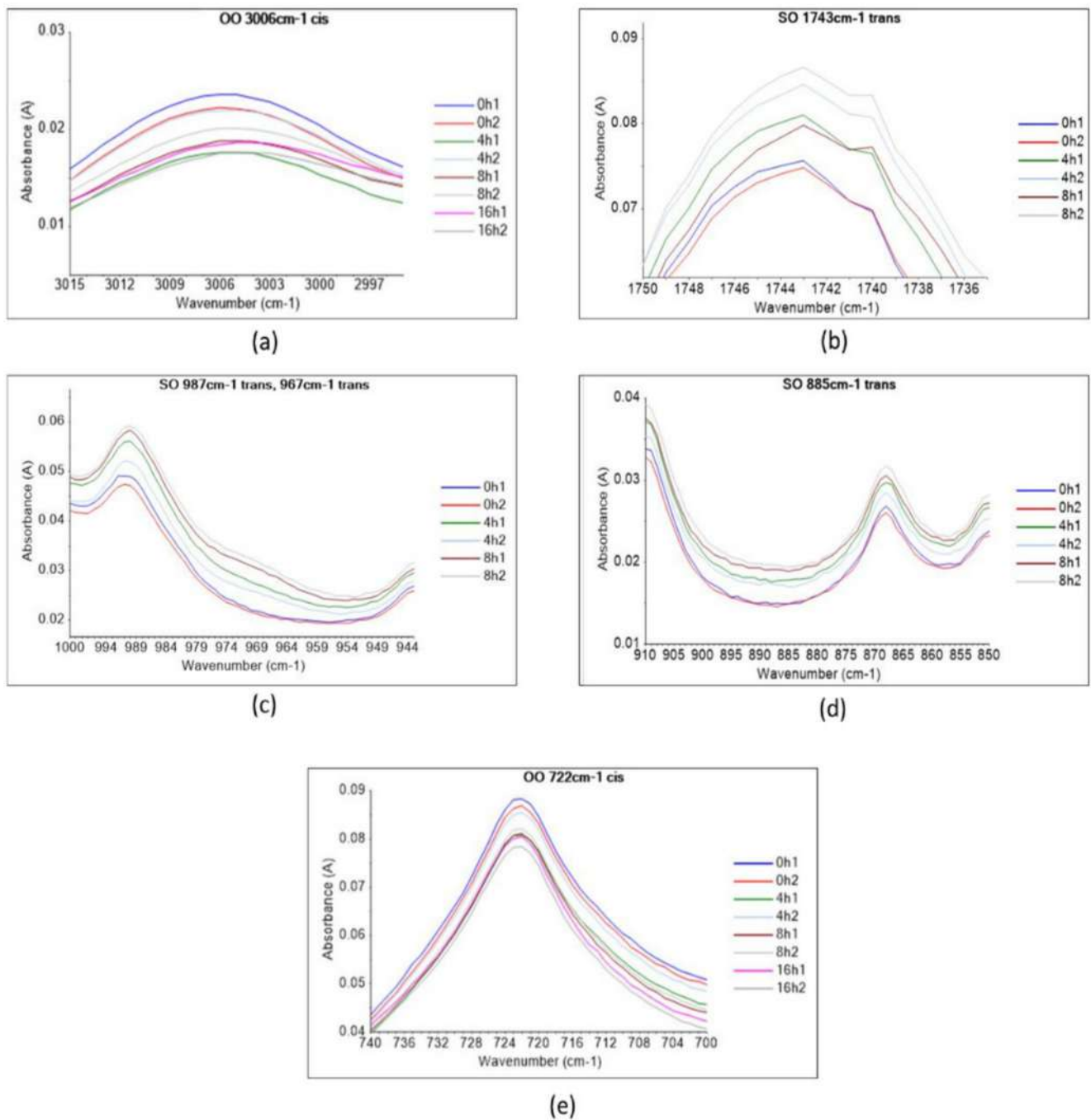
Fig. 4. The spectral changes due to heating of SO and OO samples, related to cis-trans isomerization ((a): 3006 cm^{-1} (b): 1743 cm^{-1}, (c): 987 cm^{-1} and 967 cm^{-1} d): 885 cm^{-1} and (e): 722 cm^{-1}).

Qualifying these transformations and conducting a comparative study of the thermal resistance of oils by mere visual observation is not feasible. Therefore, principle component analysis (PCA) was performed. Fig 6. Shows the score plot of PCA for the classification of SO, OO, and CO based on their resistance to cis-trans isomerization of double bonds and the formation of secondary oxidation products due to heating.
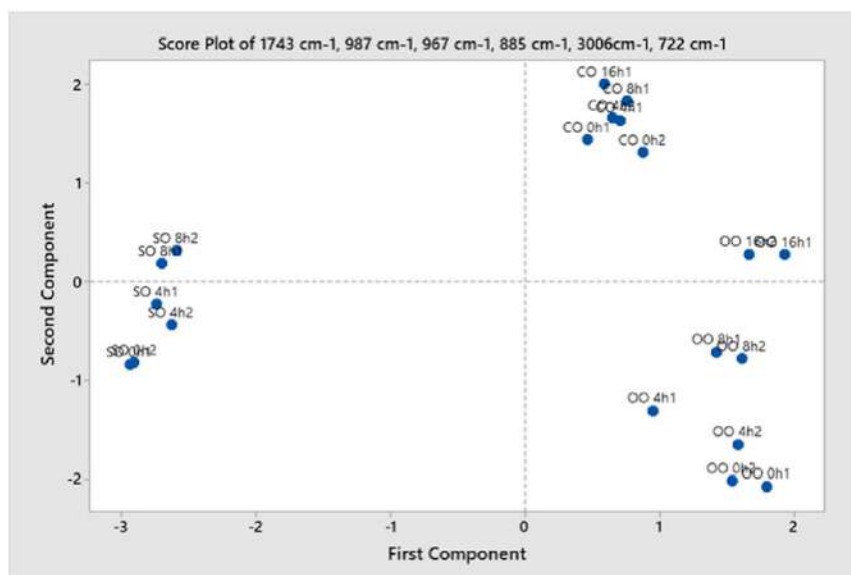
Fig. 5. PCA score plot of absorbance at 3006, 1743, 987, 967, 885 and 722 cm^{-1} for heated OO (0 - 16h), SO (0 - 8h) and CO (0 - 16h) samples

The PCA score plot in Fig. 5, developed from the spectral data of SO heated for 0-8 hours and OO and CO samples heated for 0-16 hours, accounts for approximately 88.59% of the variance using the first (PC1) and second (PC2) principal components. PC1 accounts for 60.09% of the variance, while PC2 accounts for 28.50%. The plot shows that the distance of the heated oil samples from the fresh (0-hour heated) samples increases with heating time, particularly for OO and SO samples. This indicates that the chemical composition of CO remains more similar to the fresh sample, attributed to its higher saturated fat content, which limits cis-trans isomerization, thus making it more thermally stable. These observations support the effectiveness of spectrophotometric methods in comparing the thermal stability of oils.
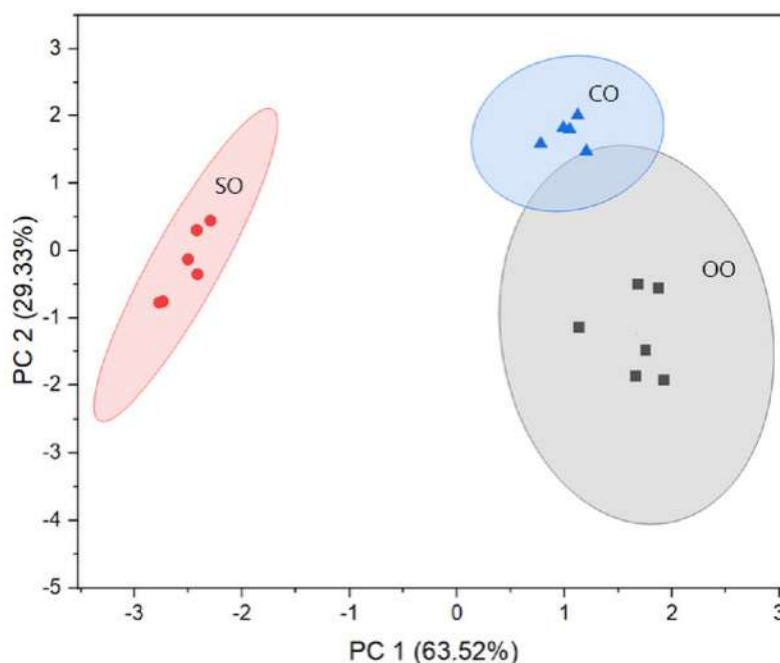

Fig. 6. PCA Cluster plot of absorbance at 3006, 1743, 987, 967, 885 and 722 cm^{-1} for 0-8h heated OO, SO and CO samples with 95% confidence level

Fig. 6. presents the PCA cluster plot with a 95% confidence level, derived from the spectral data of the three oils heated for 0-8 hours. This plot captures 92.85% of the variance using PC1 and PC2, which explain 63.52% and 29.33% of the variance, respectively. The SO samples show moderate dispersion, forming a tight cluster, while the OO samples exhibit high dispersion, forming a loose cluster. This suggests that the degree of heat-induced cis-trans isomerization and oil

oxidation in SO is less significant compared to OO. However, the 16-hour heated SO sample developed a gel-like consistency and a bright yellow color, indicating drastic deterioration after prolonged heating.
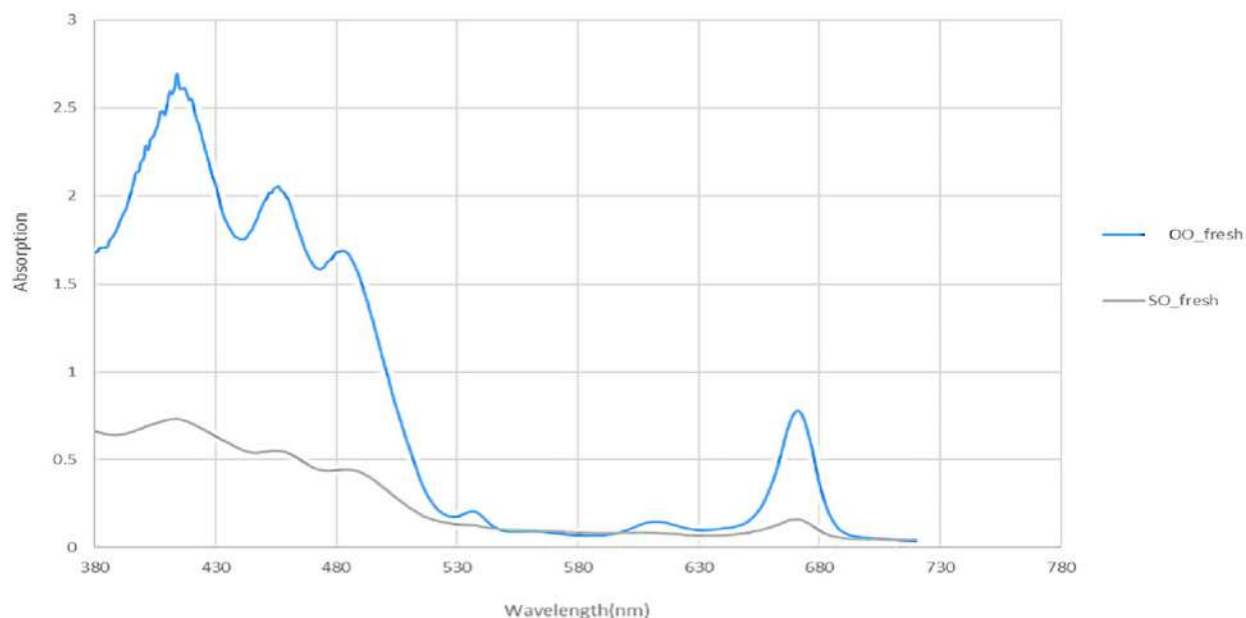
3.2 UV-Vis spectroscopy analysis

Fig. 7. Visible range absorption spectrum of Extra Virgin Olive oil and Sesame oil at room temperature.

UV-visible absorption spectra of two oils (OO and SO) showed specific criteria of recognition based on the spectra shape and absorption maxima. According to Fig. 7., both OO and SO indicate intense peaks at 417, 457, 483, and 670 nm caused by carotenoids (yellowish pigment) and chlorophyll (greenish pigment) contents. Absorption peaks at 417 nm and 670 nm were recognized as from chlorophyll a, and absorption peaks at 457 nm and 483 nm by beta carotene (Khosroshahi, 2018; Socaciu et al., 2020).
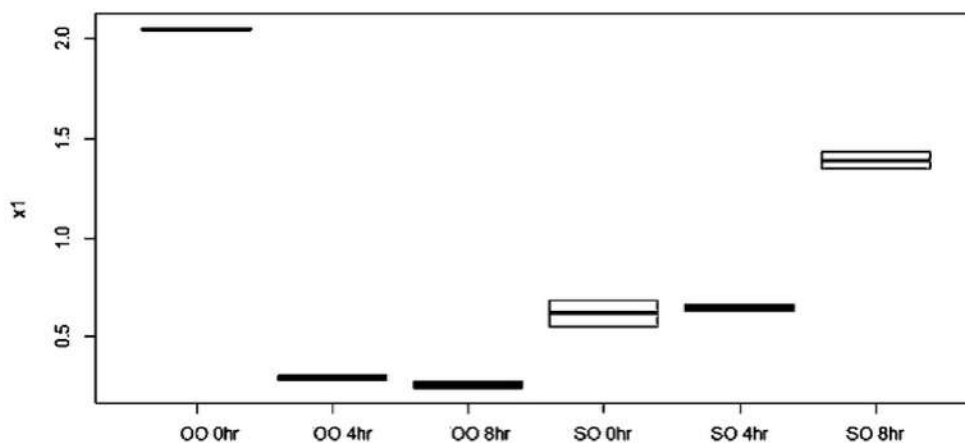

Fig. 8. Variation of the mean absorption peak at 417 nm with heating time
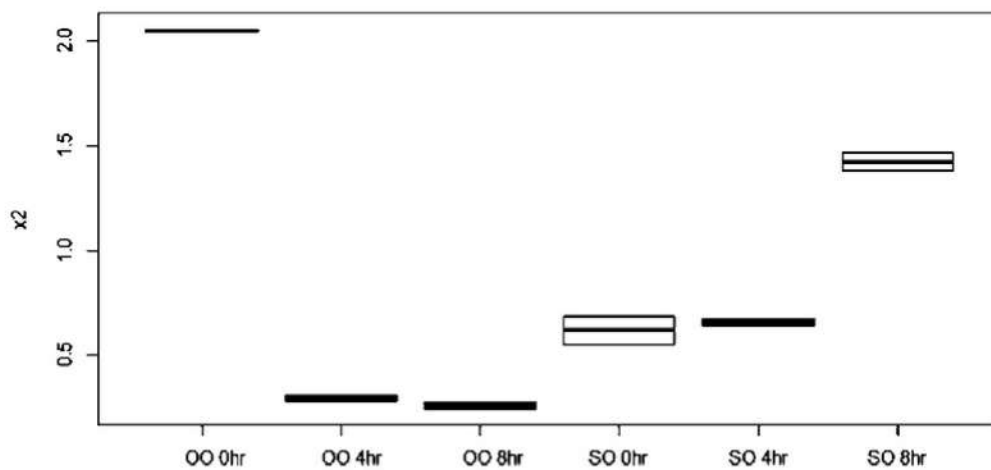
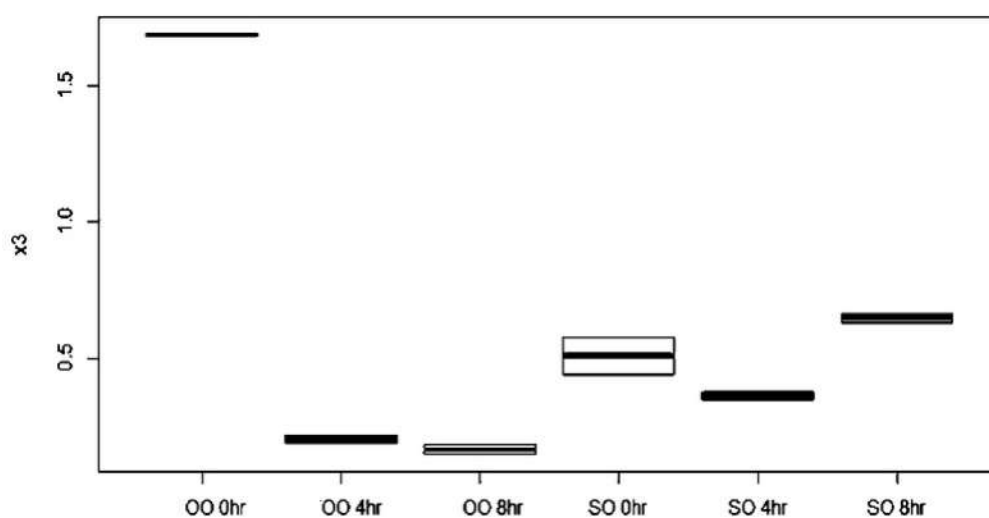**Fig. 9.** Variation of the mean absorption peak at 457 nm with heating time

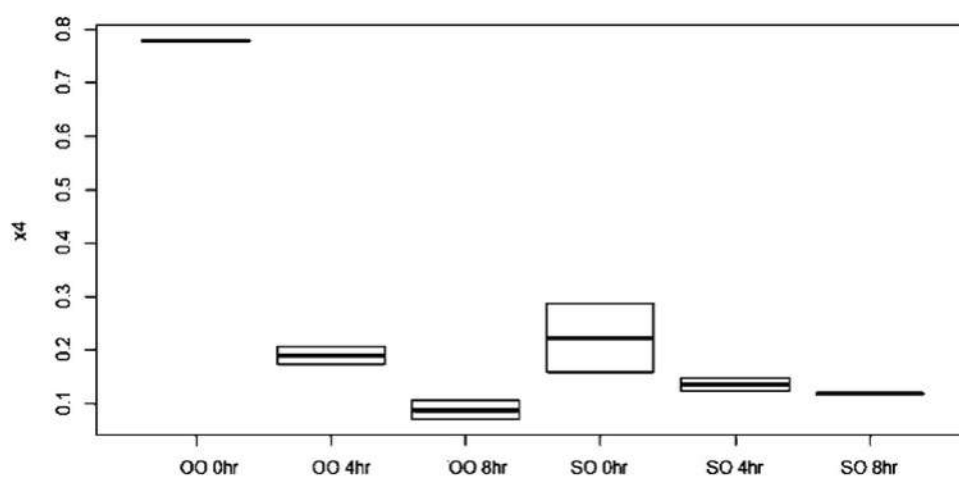**Fig. 10.** Variation of the mean absorption peak at 483 nm with heating time

Fig. 11. Variation of the mean absorption peak at 670 nm with heating time

As shown in fig 8. 9, 10, and 11, the mean absorbance of OO decreased with increasing heating time at wavelengths of 417, 457, 483, and 670 nm and it can be noted that the drop is more pronounced for OO compared to SO. Though the mean absorbance at wavelengths of 417, 457, and 483 nm for SO decreased after 4-hour heating, it can be seen that it has increased again after 8-hour heating. One reason for this is the strong oscillation in the spectra because the SO samples were not diluted.

The chlorophyll molecule is made up of a core magnesium atom surrounded by a nitrogen-containing structure known as a porphyrin ring; connected to the ring is a lengthy carbon-hydrogen side chain known as a phytol chain. During thermal processing, the central magnesium atom of chlorophyll's porphyrin ring is replaced by two hydrogen atoms to generate pheophytin, which results in a color change from bright green to brown (Weemaes et al., 1999). The boxplots represent the color change of chlorophyll a by the decrease of mean absorption at wavelengths 417 nm and 670 nm (Fig. 8 and 11). Carotenoids are made up of a polyene chain with nine conjugated double bonds and an end group at both ends. Fig. 9 and 10 show the degradation of β-carotene with increasing thermal stress as measured by a decrease in mean absorbance at 457 and 483 nm (Khosroshahi, 2018). It is not possible to quantify these alterations and undertake comparative studies of the thermal resistance of oils-based UV-visible spectra. As a result, principal component analysis (PCA) was performed. Fig. 12 exhibits a PCA score plot for the classification of OO and SO based on color variations with chlorophyll a and carotenoid deterioration as heating time increases. Principal Component 1 (PC1) accounts for 96% of the variation, whereas Principal Component 2 (PC2) accounts for 4% in the raw data from UV-visible spectra at 417, 457, 483, and 670 nm.
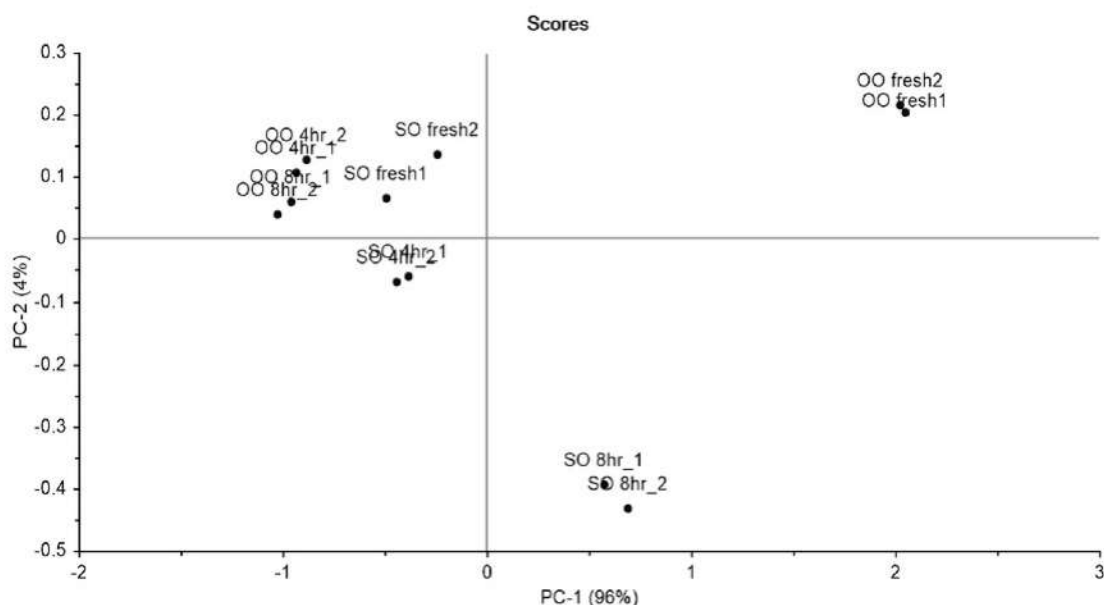

Fig. 12. PCA score plot of absorbance at wavelengths 417, 457, 483, 670 nm for SO and OO

The distance between heated samples and fresh samples illustrates the degree of color variation that has taken place due to heating. The differences in PC1 between fresh and heated OO samples are significant compared to that of SO. Thus, the increase in heating time has caused a considerable change in the chlorophyll and carotenoid content in OO. According to Fig. 12., even after 4 hours of heating, PC1 does not display any appreciable change in color in SO. Yet, the color of the SO changes substantially after 8 hours of heating. However, the color variation with increasing heating time is less pronounced in SO when compared to OO.

3.3 Free Fatty Acid

Fig. 13. depicts changes in acidity or free fatty acids (FFA). It was observed that the FFA value of SO was high compared to OO initially and that it grew considerably to reach high levels at the end of the heating time for both OO and CO.

For OO, after 16 hours of heating, the FFA composition increased from 0.29 to 1.27. After 16 hours of heating, CO's FFA content rose from 0.095 to 1.27. Though SO contains a high amount of FFA (1.95) initially, there is no significant change in FFA composition even after 16 hours of heating. It had reached 2.085 by the end of 16 hours. Thus, the FFA of OO and CO are very susceptible to changes when heated, but SO exhibits more resistance to changes in FFA caused by heating. According to the (The Gazette of the Democratic Socialist Republic of Sri Lanka, 1990), the free fatty acid (as oleic) value less than 3% is an indication for good quality SO, and less than 1% for good quality OO and CO. FFA value

of SO remains below the regulatory even after 16 hours of heating while that of OO exceeds its regulatory value after 16 hour of heating.
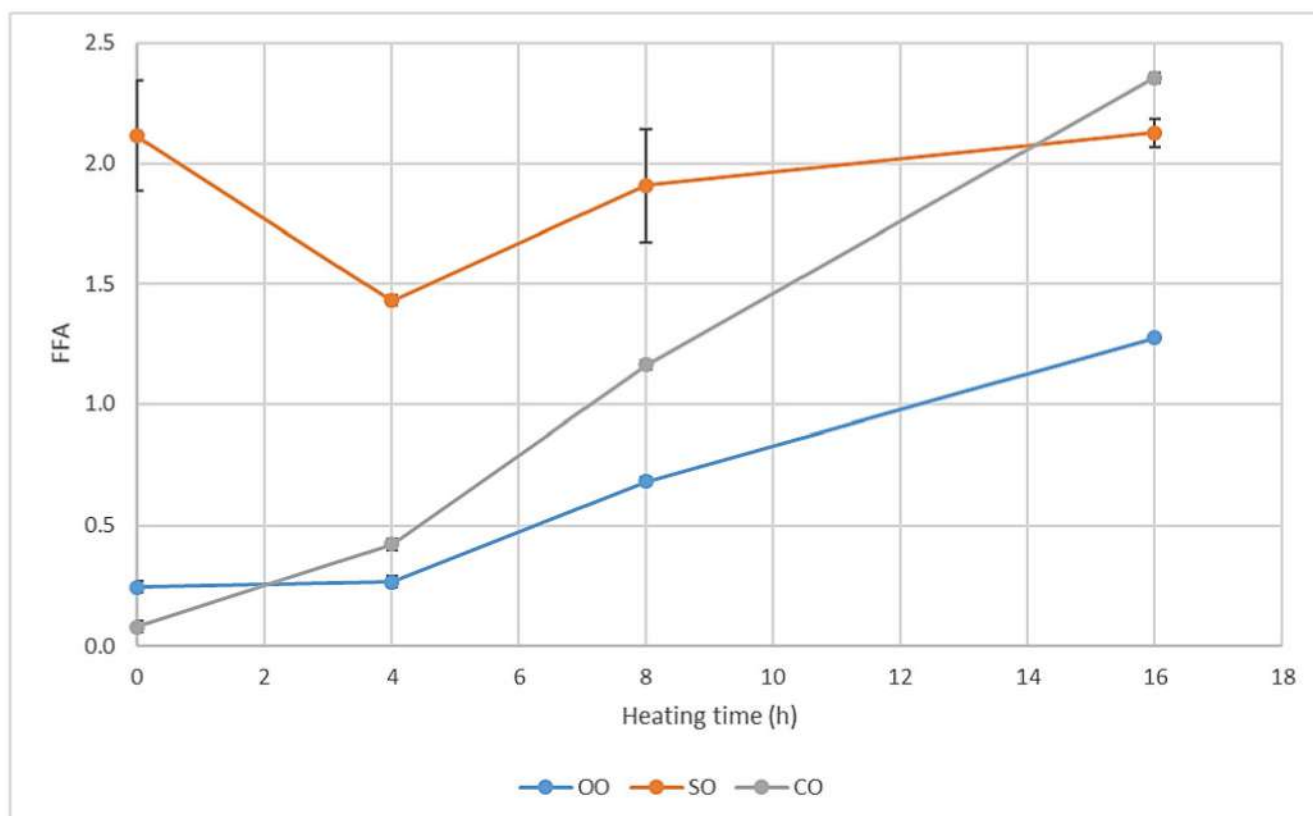

Fig. 13. Free fatty acid value with the heating time

Conclusion

The study demonstrates that FTIR and UV-visible spectroscopy, combined with PCA, effectively assess heat-induced changes in oils. Among the oils examined, SO shows superior resistance to cis-trans isomerization, color degradation, and oxidation compared to OO. However, SO significantly deteriorates after 16 hours of frying. Thus, while SO has better thermal stability than OO, it is unsuitable for extended frying periods. This underscores the importance of monitoring oil quality during extended frying processes.

Keywords: FTIR spectroscopy, UV-Vis spectroscopy, Olive Oil, Sesame Oil, Thermal Stability

References

1. Abenoza, M., De Las Heras, P., Benito, M., Oria, R., & Sánchez-Gimeno, A. C. (2016). Changes in the Physicochemical and Nutritional Parameters of Picual and Arbequina Olive Oils during Frying. *Journal of Food Processing and Preservation*, *40*(3), 353–361. https://doi.org/10.1111/jfpp.12612
2. Alves, F. C. G. B. S., Coqueiro, A., Março, P. H., & Valderrama, P. (2019). Evaluation of olive oils from the Mediterranean region by UV–Vis spectroscopy and Independent Component Analysis. *Food Chemistry*, *273*, 124–129. https://doi.org/10.1016/j.foodchem.2018.01.126
3. Borchani, C., Besbes, S., Blecker, Ch., & Attia, H. (2010). Chemical Characteristics and Oxidative Stability of Sesame Seed, Sesame Paste, and Olive Oils. *Journal of Agricultural Science and Technology*, *12*(5), 585–596. http://jast.modares.ac.ir/article-23-7694-en.html
4. Brühl, L. (2014). Fatty acid alterations in oils and fats during heating and frying. *European Journal of Lipid Science and Technology*, *116*(6), 707–715. https://doi.org/10.1002/ejlt.201300273
5. Cai, Z., Li, K., Lee, W. J., Reaney, M. T. J., Zhang, N., & Wang, Y. (2021). Recent progress in the thermal treatment of oilseeds and oil oxidative stability: A review. *Fundamental Research*, *1*(6), 767–784. https://doi.org/10.1016/j.fmre.2021.06.022

6. Casal, S., Malheiro, R., Sendas, A., Oliveira, B. P. P., & Pereira, J. A. (2010). Olive oil stability under deep-frying conditions. *Food and Chemical Toxicology*, 48(10), 2972–2979. https://doi.org/10.1016/j.fct.2010.07.036

7. Chew, S. C., & Nyam, K. L. (2019). Refining of edible oils. In *Lipids and Edible Oils: Properties, Processing and Applications* (pp. 213–241). Elsevier. https://doi.org/10.1016/B978-0-12-817105-9.00006-9

8. da Silva Oliveira, W., Neves, D. A., & Ballus, C. A. (2019). Mature chemical analysis methods for food chemical properties evaluation. In *Evaluation Technologies for Food Quality* (pp. 63–90). Elsevier. https://doi.org/10.1016/B978-0-12-814217-2.00005-6

9. Frankel, E. N. (2010). Chemistry of Extra Virgin Olive Oil: Adulteration, Oxidative Stability, and Antioxidants. *Journal of Agricultural and Food Chemistry*, 58(10), 5991–6006. https://doi.org/10.1021/jf1007677

10. Gandul-Rojas, B., & Minguez-Mosquera, M. I. (1996). Chlorophyll and Carotenoid Composition in Virgin Olive Oils from Various Spanish Olive Varieties. *Journal of the Science of Food and Agriculture*, 72(1), 31–39. https://doi.org/10.1002/(SICI)1097-0010(199609)72:1<31::AID-JSFA619>3.0.CO;2-5

11. Gonçalves, R. P., Março, P. H., & Valderrama, P. (2014). Thermal edible oil evaluation by UV-Vis spectroscopy and chemometrics. *Food Chemistry*, 163, 83–86. https://doi.org/10.1016/j.foodchem.2014.04.109

12. Guille, M. D., & Cabo, N. (2002). Analytical, Nutritional and Clinical Methods Section Fourier transform infrared spectra data versus peroxide and anisidine values to determine oxidative stability of edible oils. *Food Chemistry*, 77(4).

13. Karamizadeh, S., Abdullah, S. M., Manaf, A. A., Zamani, M., & Hooman, A. (2013). An Overview of Principal Component Analysis. *Journal of Signal and Information Processing*, 04(03), 173–175. https://doi.org/10.4236/jsip.2013.43B031

14. Khosroshahi, M. E. (2018). Effect of Temperature on Optical Properties of Vegetable Oils Using UV-Vis and Laser Fluorescence Spectroscopy. *Optics and Photonics Journal*, 08(07), 247–263. https://doi.org/10.4236/opj.2018.87021

15. Laguerre, M., Lecomte, J., & Villeneuve, P. (2007). Evaluation of the ability of antioxidants to counteract lipid oxidation: Existing methods, new trends and challenges. In *Progress in Lipid Research* (Vol. 46, Issue 5, pp. 244–282). https://doi.org/10.1016/j.plipres.2007.05.002

16. Li, C., Zhang, Y., Li, S., Wang, G., Xu, C., Deng, Y., & Wang, S. (2013). Mechanism of formation of trans fatty acids under heating conditions in triolein. *Journal of Agricultural and Food Chemistry*, 61(43), 10392–10397. https://doi.org/10.1021/jf402854b

17. Nielsen, S. S. (2017). *Food Analysis Laboratory Manual*. Springer International Publishing. https://doi.org/10.1007/978-3-319-44127-6

18. Omar, M., Hazwani, M., Nazreen, M., & Zuberdi, A. (2014). Studies on Frying Quality of Virgin Coconut Oil and Shortening Blends. *Oriental Journal of Chemistry*, 30(3), 1279–1286. https://doi.org/10.13005/ojc/300344

19. Poiana, M. A., Alexa, E., Munteanu, M. F., Gligor, R., Moigradean, D., & Mateescu, C. (2015a). Use of ATR-FTIR spectroscopy to detect the changes in extra virgin olive oil by adulteration with soybean oil and high temperature heat treatment. *Open Chemistry*, 13(1), 689–698. https://doi.org/10.1515/chem-2015-0110

20. Poiana, M. A., Alexa, E., Munteanu, M. F., Gligor, R., Moigradean, D., & Mateescu, C. (2015b). Use of ATR-FTIR spectroscopy to detect the changes in extra virgin olive oil by adulteration with soybean oil and high temperature heat treatment. *Open Chemistry*, 13(1). https://doi.org/10.1515/chem-2015-0110

21. Rohman, A., & Che Man, Y. B. (2013). Application of FTIR Spectroscopy for Monitoring the Stabilities of Selected Vegetable Oils During Thermal Oxidation. *International Journal of Food Properties*, 16(7), 1594–1603. https://doi.org/10.1080/10942912.2011.603874

22. Sharanke, K., & Sivakanthan, S. (2022). Comparative study on stability of coconut oil, sunflower oil and palm oil during continuous deep frying. *Ceylon Journal of Science*, 51(4), 359. https://doi.org/10.4038/cjs.v51i4.8053

23. Socaciu, C., Fetea, F., Ranga, F., Bunea, A., Dulf, F., Socaci, S., & Pintea, A. (2020). Attenuated total reflectance-fourier transform infrared spectroscopy (Atr-ftir) coupled with chemometrics, to control the botanical authenticity and quality of cold-pressed functional oils commercialized in romania. *Applied Sciences (Switzerland)*, 10(23), 1–16. https://doi.org/10.3390/app10238695

24. Sunisa W., Worapong U., Sunisa S., Saowaluck J., & Saowakon W. (2011). Quality changes of chicken frying oil as affected of frying conditions. *International Food Research Journal*.

25. *The Gazette of the Democratic Socialist Republic of Sri Lanka*. (1990).

26. Tótoli, E. G., & Salgado, H. R. N. (2015). A green approach for the quantification of daptomycin in pharmaceutical formulation by UV spectrophotometry. *Brazilian Journal of Pharmaceutical Sciences*, 51(4), 811–821. https://doi.org/10.1590/S1984-82502015000400007

27. Wan, Y., Li, H., Fu, G., Chen, X., Chen, F., & Xie, M. (2015). The relationship of antioxidant components and antioxidant activity of sesame seed oil. *Journal of the Science of Food and Agriculture*, 95(13), 2571–2578. https://doi.org/10.1002/jsfa.7035

28. Weemaes, C. A., Ooms, V., van Loey, A. M., & Hendrickx, M. E. (1999). Kinetics of chlorophyll degradation and color loss in heated broccoli juice. *Journal of Agricultural and Food Chemistry*, 47(6), 2404–2409. https://doi.org/10.1021/jf980663o

29. Yu, K. S., Cho, H., & Hwang, K. T. (2018). Physicochemical properties and oxidative stability of frying oils during repeated frying of potato chips. *Food Science and Biotechnology*, 27(3), 651–659. https://doi.org/10.1007/s10068-017-0292-y