African Journal of Advanced Pure and Applied Sciences (AJAPAS)
Online ISSN: 2957-644X
Volume 4, Issue 1, January - March 2025
Page No: 233-237
Website: https://aaasjournals.com/index.php/ajapas/index
ISI 2024: 0.877 **SJIFactor 2024: 6.752** **معامل التأثير العربي: 1.62**

Determination the acidity of olive oil samples stored at different periods of times in Tarhouna, Libya

Mahmud Ashtewi S. Ashtewi [1 *], Salem Mohammed S. Ahmed [2]
[1,2] Department of Chemistry, Faculty of Science, Azzaytuna University, Tarhouna, Libya

تقدير درجة الحموضة لعينات من زيت الزيتون مخزنة لفترات زمنية مختلفة في منطقة ترهونة، ليبيا

محمود اشتيوي صالح اشتيوي [1 *]، سالم محمد سالم أحمد [2]
[2،1] قسم الكيمياء، كلية العلوم، جامعة الزيتونة، ترهونة، ليبيا

*Corresponding author: mah_2004_mod@yahoo.com

Received: November 20, 2024	Accepted: January 30, 2025	Published: February 09, 2025

Abstract:
Random olive oil samples from local farmers in Tarhouna stored at different periods were investigated to draw a conclusion about their influence with period storage. Ten samples of virgin olive oil stored at different periods (ranged between few months to more than fifteen years) were collected and pre-handled for analysis. The acidity of the samples was determined by volumetric titration using KOH according to the standard BS EN ISO 660 2009. The obtained results showed irregular increase in acidity for samples aged less than three years, while samples aged more than three years showed fairly steady increase in acidity. The study found that the samples stored for more than three years have acidity values exceed codex Alimentarius of olive oil (3.3%), while the samples aged for less than three years have acidity values varied around 3.3%, which may be attributed to poor harvest and pre-extraction procedures, as well as to long storage time.

Keywords: olive oil, harvest, storage period, oil acidity, volumetric and KOH.

الملخص
أجريت هذه الدراسة لقياس درجة حموضة عينات من زيت الزيتون المُخزنة لفترات زمنية مختلفة، لمعرفة مدى تأثرها بمدة التخزين. تم جمع 10 عينات عشوائية من زيت الزيتون البكر ذات فترات تخزين مُختلفة (تراوحت ما بين عدة أشهر إلى أكثر من 15 سنة). تم قياس درجة الحموضة بطريقة المعايرة الحجمية باستعمال هيدروكسيد البوتاسيوم KOH وفقاً للطريقة القياسية المُعتمدة BS EN ISO 660 2009. وقد أظهرت النتائج المُتحصل عليها زيادة غير مُنتظمة في درجة الحموضة في العينات ذات فترات التخزين الأقل من 3 سنوات، في حين أظهرت نتائج التحليل زيادة شبه منتظمة في درجة الحموضة للعينات المُخزنة لأكثر من 3 سنوات. وقد توصلت الدراسة إلى أن درجة الحموضة في العينات المُخزنة لأكثر من 3 سنوات تتجاوز الحد المسموح به حسب الدليل الغذائي والمُقدر بحوالي 3.3%، في حين تراوحت درجة حموضة عينات زيت الزيتون في العينات المُخزنة لفترة تقل عن 3 سنوات حول القيمة 3.3%، وقد أرجع سبب ذلك إلى عدة أسباب منها عدم اتباع الخطوات الصحيحة أثناء عمليات القطاف والخطوات التي تسبق عمليات عصر الزيتون، إضافة إلى طول مدة التخزين.

الكلمات المفتاحية: زيت الزيتون، عمليات القطاف، مدة التخزين، حموضة الزيت، الطريقة الحجمية، KOH.

Introduction

The olive tree is one of the oldest known cultivated trees in the world [1,2]. It grows in a wide range of soils and different environmental conditions. It is essential to almost all Mediterranean countries and easily adaptive to their environment. Libya is one of the Mediterranean countries that locates at the north coast of Africa. The production of olive oil can be carried out by cold pressing of fresh and healthy olive fruits or by other physical processes under low thermal conditions. The olive oil in Libya is one of the culture heritage for all people. In Libya, olive

oil is produced by privet manufacturers using cold pressing [3, 4]. The average production of olive oil in Libya is about 0.5% of the total amount produced over the world [3,5].
The characteristic aroma of the olive oil makes it unique among other vegetable oils, such characteristic is ascribed to its balanced composition of fatty acids and other minor components (such as phenolic compounds, alpha-tocopherol, carotenoids, squalene, simple triterpenes and volatile compounds). In comparison with other seed oils that contain high amounts of polyunsaturated fatty acids, olive oil is predominated by monounsaturated fatty acids, which are less subject to oxidation, therefore, its stability and expiration time is increased [6-8]. The main two monounsaturated fatty acids are oleic acid and palmitoleic acid, such acids are beneficial to human health especially for people who have metabolic syndromes, diabetes or other health problems. Studies have revealed that these fatty acids have critical role in decreasing the total cholesterol and **low-density lipoprotein** (LDL cholesterol). When monounsaturated fatty acids replace saturated fatty acids in human body, they reduce triglycerides and increase or maintain the level of **high-density lipoprotein** (HDL cholesterol). Moreover, the olive oil provides primary and secondary protection against developing of cardiovascular diseases [9].
Olive oils have been classified mainly into three common categories; these are virgin olive oil, refined olive oil, and olive pomace oil. Each has its unique mill method, composition, flavour characteristics, and particular nutritional uses. Virgin olive oil is obtained by milling fresh olives, followed by automated extraction of the oil. This process is conducted without heat or chemicals. In order to increase the quality olive oil, the fruit must be collected, harvested, and stored in a right manner. This category of olive oil has high flavourful and health-enhancing [10, 11]. However, refined olive oil is produced from low quality of olive oil that experiences to a refining process to remove the remaining of the free fatty acids (FFA) and other impurities. The FFA are an undesirable component of oils. On the other hand, beneficial substances, such as polyphenols, are also removed during the refining process. As a result, the obtained oil is colourless, odourless and tasteless [10,12]. While, olive pomace oil is extracted from olive mash using chemical solvents at high temperature [10].
Virgin olive oil is the main category of olive oil in Libyan society [13]. It has moderate flavour, odour and slightly low free fatty acid content [10]. However, the quality of olive oil is influenced by several of factors, such as irrigation and fertilization (agronomic factors), harvesting and maturity (cultivation practices), post-harvest storage and extraction system (technological factors) [14 – 17]. FFA are one of the major reasons that shorting shelf life and flavour deterioration of oil [18, 19]. FFA are considered as the main parameters used to evaluate the quality of olive oil. The content of FFA from oleic acid is used to determine the category of olive oil. According to the International Olive Oil Council, virgin olive oil has a maximum of 3.3 % FFA, such category is extracted from olive fruits mechanically without any thermal or chemical treatment. [18, 20]. Many studies have been carried out to investigate effects of air and sunlight exposure [21], storage time and type of containers [22 - 24] on the acidity of olive oil and therefore on the quality of oil. The results showed a gradual loss of oil quality with increasing FFA contents when oils stored in plastic or glass bottles [22]. Other study found that the acidity increases incrementally as a function of storage time [23].
The olive oil produced in Libya is obtained by cold pressing and stored at different conditions for long period, as well as it may be influenced by several factors including harvesting, cultivation practices, post-harvest, extraction system and types of containers. All these factors may affect the acidity of olive oil. Therefore, this study aims to determine the acidity of olive oil in random samples in Tarhouna at different storage periods to investigate the oil acidity and its sources.

Material and methods

Samples collections

The study was carried out on ten random olive oil samples obtained from olive fruits harvested from different local privet farmers in Tarhouna, Libya. The rain-fed olive fruits were hand-picked, collected at different seasons, and stored in nets or sacks at ambient temperatures for different times until their pressing. The extraction process of olive oil was conducted with different cold oil-press. Olive oil samples were stored in various containers without ideal conditions.

Preparation and Standardization of Reagents

All chemicals were analytical grade and prepared according to procedures in the standard [25, 26]. *Phenolphthalein indicator* was prepared by weighting 1.0 g of phenolphthalein and dissolved in 100 mL of ethanol. *Potassium hydroxide titrant* was prepared by accurately weighting 1.4 g of potassium hydroxide and place it in a 250-mL volumetric flask. Make up to the mark with water. A mixture of *Ethanol- diethyl ether solution* (1:1, v/v) was prepared by mixing equal volumes.
Standardization of potassium hydroxide titrant. 0.21 g of potassium hydrogen phthalate was accurately weighted, previously dried to constant weight at 105 ^{o}C, and dissolved in 50 mL of distilled water in a 250-mL conical flask and shook well. Then two drops of phenolphthalein indicator were added and the solution titrated with the potassium hydroxide titrant until pink coloration can be observed. The exact concentration of the potassium hydroxide titrant was calculated according to the following equation:

$$C_{KOH} = \frac{W_{C_8H_5KO_4} \times 1000}{V_{KOH} \times Mw_{C_8H_5KO_4}}$$

where C_{KOH}= Molarity of potassium hydroxide titrant (mol/L), V_{KOH} = Volume of potassium hydroxide titrant used (mL), $Mw_{C_8H_5KO_4}$= Molecular weight of potassium hydrogen phthalate (204.22 g/mol), $W_{C_8H_5KO_4}$ = Weight of potassium hydrogen phthalate used (g).

Titration of oil samples

1.0 g of olive oil sample was accurately weighted and placed in a 250-mL conical flask. 20 mL of ethanol-ether mixture was then added to the oil sample and shook well. The solution was titrated with potassium hydroxide titrant until pink color can be observed which persists for 30 sec. Finally, the volume of potassium hydroxide titrant used was measured and the acid value calculated according to the following equation:

$$\text{acid value} = \frac{V_{KOH} \times M_{KOH} \times 56.1}{W}$$

Where V_{KOH} = Volume of potassium hydroxide titrant used (mL), M_{KOH} = molarity of potassium hydroxide (mole/L), W = Weight of the fatty oil being examined (g), 56.1 = Molecular weight of KOH (g/mol).
The acidity is frequently expressed as free fatty acid for which calculation will be:

$$\text{Free fatty acids (as \% oleic acid)} = \frac{V_{KOH} \times M_{KOH} \times 28.2}{W}$$

Where, 28.2 = Molecular weight of oleic acid divided by 10.
Acid value = Percent fatty acid (as oleic) × 2.0

Results and discussion

The free fatty acids FFA (as % oleic acid) and acid values (AV) for olive oil samples were calculated and listed in the table 1, and shown in figure 1.

Table (1): The free fatty acids FFA and acid values (AV) for collected olive oil samples.

Sample's No.	Storage period (years)	Volume of KOH (mL)	Fatty acids percentage (%)	Acid value (mg KOH/g oil)
1	< 1	0.60	1.65	3.30
2	< 1	1.60	4.40	8.80
3	≈1	4.70	12.92	25.84
4	≈1	1.00	2.75	5.50
5	≈3	2.40	6.60	13.20
6	≈3	1.40	3.85	7.70
7	≈6	3.80	10.45	20.90
8	≈10	2.80	7.70	15.40
9	≈15	4.10	11.27	22.54
10	> 15	9.80	26.94	53.88

Exact molarity of KOH = 0.098 M

The results listed in table 3 illustrated fairly increasing FFA values in the sample with time which is in agreement with literature [22- 24]. However, few variations in FFA were observed in samples aged less than 3 years, which may be attributed to several factors such as harvesting and maturity of fruits, post-harvest storage and extraction system.

The obtained results obviously showed marked increasing in FFA with time (26.94%) for the oil sample stored more than 15 years. However, the results showed also that the oil sample that stored for only one year has high FFA up to 12.92%, which is unexpected for such short period of time which may be attributed to poor harvesting and post-harvesting handling. Additionally, there is an obvious variation between the two samples that aged for similar period of time (one years) after extraction; one of which has FFA equal to 1.65%, and the other has 4.40% of FFA which are fairly different for two samples aged for the same period. The similar phenomenon was observed also for the two samples stored for three years after extraction (6.60% and 3.85%). Such spikes in acidity values

for the two samples stored at same period of time might lead us to conclude an interesting finding. These variations in acidity for the same period of time may be due to handling and post-harvest process of olive fruits.

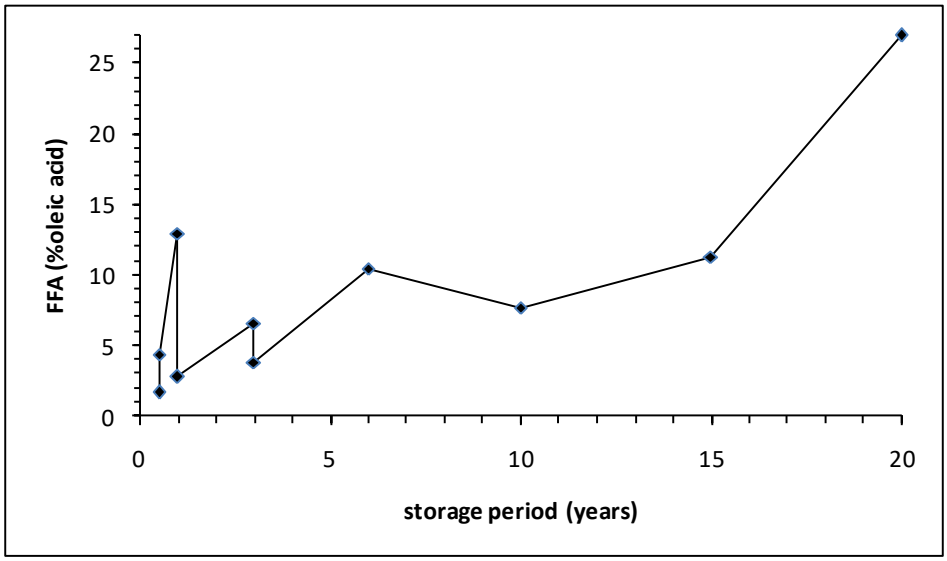

Figure (1): Effect of storage period on acidity of olive oil samples.

By comparing the obtained results in this study with codex standard for olive oil, it is found that most of the samples exceed the criterion specified by codex alimentarius (3.3%), with exception of two samples that stored for less than one year. Such observations are a compatible with literatures that stated that acidity of oils increases with storage time after pressing olive fruits [22- 24]. Moreover, it is noted that the acidity of the samples stored for more than three years were not steady increased which may be attributed to unsound harvest procedures. However, two of oil samples whose acidities are 4.40% (stored less than one year) and 12.92% (stored around one year) exhibit marked elevated acidities which exceed codex alimentarius 3.3%. These two values reveal poor harvesting procedures of olive fruits on that season as well as poor storing conditions such as type of containers, extent sun exposure and ambient temperature.

In general, the obtained results illustrated that most of the samples have elevated content of FFA. Such observations revealed that harvest and pre-extraction operations may be unsatisfactory, and there are several factors led to increase the acidity of the olive oil samples. Among these factors, limited oil extraction capacities of the industrial facilities, which leads to slow processing of olive fruits post-harvest and, in turn, leads to piled harvested olives in heaps for up to several weeks before pressing the olive fruits. Also pressure within the olive pile can damage the fruits and the fluid secretion from crushed olives may provide an optimum media for growth of fungi and bacteria. Numerous studies [8,16,17,18]. have been revealed that the storing olive fruits in silos, nets or sacks for period of time lead to an increase enzymatic breakdown of the olive cell structure, especially if the fruits had been battered during harvesting. Also the contact between olive and water a fluidic waste resulting from olive oil extraction is one of the factors that increase FFA in olive oil. Furthermore, biding olive fruits at ambient temperature before extraction has been proven to increase the oil acidity [27].

Conclusion

An overall conclusion can be deduced from this study is that olive oil samples stored for more than 3 years exceed of codex alimentarius (> 3.3%) and not recommended for human consumption, while samples that stored for one year or less have fairly low content of FFA. The study also concluded that elevated values for all olive oil samples may be mainly ascribed to process that precede pressing olive fruits such as harvesting and maturity of fruits, post-harvest storage and extraction system, as well as to aging period.

References

[1]- Liphschitz N, Gophna R, Hartman M and Biger G. The beginning of olive (Olea europaea) cultivation in the old world: a re-assessment. *J Arch Sci* (1991) 18:441–453

[2]- Iosif Emmanouil Kapellakis, Konstantinos P. Tsagarakis and John C. Crowther. Olive oil history, production and by-product management. *Rev Environ Sci Biotechnol* 2008 7:1–26.

[3]- Vesna B. Vujasinović1, Biljana B. Rabrenović, Seddiq M. A. Esalami, Sanja B. Dimić, Dragan V. Tešanović and, Maja V. Banjac. Sensory and some chemical characteristics of olive oils produced in Libya. *1st International Conference on Advanced Production and Processing* – ICAPP 2019 Novi Sad, Serbia, October 10-11, 2019.

[4]- NEPAD–CAADP Bankable Investment Project Profile. *Food Security Scheme* (Wheat, Dates & Olives, Seed Production). Volume II 2006.

[5]- IOOC, 2014. World Olive Oil Figures – Production. http://www.internationaloliveoil.org/estaticos//view/131-world-olive-oil-figures. Accessed February 20, 2019
[6] - Mina Kafkaletou, Georgia Ouzounidou and Eleni Tsantili. Fruit Ripening, Antioxidants and oil composition in koroneiki olives (Olea europea L.) at different maturity indices. *Agronomy* 2021, 11, 122. https://doi.org/10.3390/agronomy11010122.
[7]- Conde, C., Delrot, S. and Gerós, H. Physiological, biochemical and molecular changes occurring during olive development and ripening. *J. Plant Physiol.* 2008, 165, 1545–1562. [CrossRef] [PubMed]
[8]- Sanchez, J. and Harwood, J. Biosynthesis of triacylglycerols and volatiles in olives. *Eur. J. Lipid Sci. Technol.* 2002, 104, 564–573. [CrossRef]
[9]- Salazar DM, López-cortés I and Salazar-garcía DC. Olive oil: Composition and health benefits. Editors: T. Fritjof and B. Henning © 2017 nova science publishers, inc, 1.
[10]- Wendy J. Dahl, Michael A. Tandlich and Julie England. Health benefits of olive oil and olive extracts. *Food science and human nutrition*, FSHN16-4. 2022.
[11]- Buckland G and Gonzalez CA. The Role of olive oil in disease prevention: A Focus on the recent epidemiological evidence from cohort studies and dietary intervention trials. *Br J Nutr* (2015) 113:S94–101. doi: 10.1017/S0007114514003936 - DOI - PubMed.
[12] - Institute of Shortening and Edible Oils (ISEO). Food fats and oils. Washington, DC: Institute of Shortening Fats and Oils. http://www.iseo.org/httpdocs/FoodFatsOils 2016. pdf.
[13]- A. A. Allaq, N. J. Sidik, A. Abdul-Aziz, A. Elengoe, H. M. Agha, A. I. Abdulrazzaq and F. Kouider. Overview of the character analysis of libyan olive oil and their advantages in biomedical applications. *Journal of Asian Scientific Research*, vol. 12, pp. 53-61, 2022.Available at: https://doi.org/10.55493/5003.v12i1.4474.
[14]- El Qarnifa, S., El Antari, A., and Hafidi, A. Effect of maturity and environmental conditions on chemical composition of olive oils of introduced cultivars in morocco. *Journal of Food Quality*, 2019, 1–14. https://doi.org/10.1155/2019/1854539.
[15]- O. Baccouri, M. Guerfel, B. Baccouri et al. Chemical composition and oxidative stability of Tunisian monovarietal virgin olive oils with regard to fruit ripening. *Food Chemistry*, vol. 109, no. 4, pp. 743–754, 2008.
[16]- A. Dag, Z. Kerem, N. Yogev, I. Zipori, S. Lavee, and E. Ben-David. Influence of time of harvest and maturity index on olive oil yield and quality. *Scientia Horticulturae*, vol. 127, no. 3, pp. 358–366, 2011.
[17]- A. Rotondi, A. Bendini, L. Cerretani, M. Mari, G. Lercker, and T. G. Toschi. Effect of olive ripening degree on the oxidative stability and organoleptic properties of cv. Nostrana di Brisighella extra virgin olive oil. *Journal of Agricultural and Food Chemistry*, vol. 52, no. 11, pp. 3649–3654, 2004.
[18]- El-Abassy R.M., Donfack P. and Materny A. Rapid determination of free fatty acid in extra virgin olive oil by Raman spectroscopy and multivariate Analysis. *J Am Oil Chem Soc* **86**, 507–511 (2009). https://doi.org/10.1007/s11746-009-1389-0.
[19]- Perretti G, Finotti E, Adamuccio S, Della Sera R and Montanari L. Composition of organic and conventionally produced sunflower seed oil. J Am Oil Chem (2004) Soc 81:1119–1123.
[20]- O'Brien RD. Fats and oils, formulating and processing for applications, 2nd edn. CRC Press, *Boca Raton*, (2004) pp 34–35.
[21]- Orwa Jaber Houshia, Mohamad abuEid, Oday Zaid, Hazem Shqair, Motasem Zaid, Wala Nashariti, Batool Noor and Fuad Al-Rimwai. Alteration of Nabali Baladi Extra Virgin Olive Oil (EVOO) chemical parameters as a function of air and sunlight exposure. OCL, 26 (2019) 38 DOI: https://doi.org/10.1051/ocl/2019036.
[22]- Méndez AI and Falqué E. Effect of storage time and container type on the quality of extra-virgin olive oil. *Food Control*. 2007; 18, 521-529. https://doi.org/10.1016/j.foodcont.2005.12.012.
[23]- Nierat, T.H. Storage age dependence of olive oil acid in different locations in palestine. Article *in Journal of Physical Therapy Science* · January 2013. https://www.researchgate.net/publication/265595026
[24]- Ghanbari Shendi E, Sivri Ozay D, Ozkaya MT and Ustunel NF. Changes occurring in chemical composition and oxidative stability of virgin olive oil during storage. OCL, 2018; 1-8. https://doi.org/10.1051/ocl/2018052.
[25]- BS EN ISO 660 2009. Animal and vegetable fats and oils. – Determination of acid value and acidity. *British Standard*.
26- Official and Tentative Methods of the American Oil Chemist Society, Method Cd 3d 63, American Oil Chemists Society, Champaign 1989.
[27]- Dag, A., Boim, S., Sobetin, Y. and Zipori, I. Effect of mechanically harvested olive storage temperature and duration on oil quality. *Horttechnology* 2012, *22*, 528–533, doi:10.21273/HORTTECH.22.4.528.