Accepted Manuscript

Fresh extra virgin olive oil, with or without veil

José Antonio Cayuela Sánchez, Belén Caballero Guerrero

PII: S0924-2244(18)30481-3

DOI: https://doi.org/10.1016/j.tifs.2018.11.014

Reference: TIFS 2359

To appear in: *Trends in Food Science & Technology*

Received Date: 12 July 2018

Revised Date: 8 November 2018

Accepted Date: 8 November 2018

Please cite this article as: Cayuela Sánchez, J.A., Guerrero, B.C., Fresh extra virgin olive oil, with or without veil, *Trends in Food Science & Technology*, https://doi.org/10.1016/j.tifs.2018.11.014.

Fresh extra virgin olive oil, with or without veil

José Antonio Cayuela Sánchez[1], Belén Caballero Guerrero[2]

[1]*Department of Quality and Characterization of Lipids, Instituto de la Grasa, CSIC*

[2]*Unit of Microbiology. Department of Food Biotechnology, Instituto de la Grasa, CSIC*

Abstract

Background: Fresh virgin olive oils *(VOO) constitute a complex* ***colloid*** *emulsion-sol. Their features are different to the product in which they convert after filtering or resting a time in the storage tanks. Several studies have indicated the oxidative stability of* ***unfiltered*** *VOO is better preserved than that in the filtrate ones. However, this issue is controversial. Besides, the existing knowledge on* ***veiled olive oils*** *is little.*

Scope and Approach: *This review aims to help clarify essential aspects of veiled VOO, little known, as well as raise a novel consideration on these products.*

Key Findings: *Fresh VOO veiled are not generally managed as perishable products. The new concept* ***'colloidal stability'*** *is suggested to characterize the time the colloidal dispersion last in fresh veiled VOO. The relationships between the stabilities colloidal and oxidative of veiled VOO are not well known. In complementary research presented in this review, important differences between water inclusions, formed when artificially adding water to VOO, and the* ***colloidal water bags*** *characteristic of veiled VOO, have been revealed. An internal content has been shown in this lat.*

Conclusions: *The role of the different components of fresh veiled EVOOs on the colloidal stability is far from being well known. The physiochemical state of veiled VOO may explicate the seeming contradictions on benefits from its dispersed matter. The hypothesis that* ***amphiphilic molecules*** *present in veiled VOO may locate at the interface of colloidal water bags, is suggested. They likely may form a* ***pseudo-layer****, protecting the inner content to some extent.*

Keywords: *colloid; colloidal stability; fresh; unfiltered; virgin olive oil; veil.*

Fresh extra virgin olive oil, with or without veil

José Antonio Cayuela Sánchez[1], Belén Caballero Guerrero[2]

[1]*Department of Quality and Characterization of Lipids, Instituto de la Grasa, CSIC*

[2]*Unit of Microbiology. Department of Food Biotechnology, Instituto de la Grasa, CSIC*

Fresh or Integral Virgin Olive Oil

Virgin olive oils (VOO) have different features when fresh to the product in which they convert after a few weeks or months standing in the storage tanks. This quality of the fresh VOO is visible to the human eye as a veiled appearance, due to the matter scattered in them. These VOO constitutes a colloid or dispersion (Xenakis, Papadimitriou, & Sotiroudis, 2010; Papadimitriou et al., 2013), referred to in the literature as veiled, opalescent, cloudy or integral. The colloidal dispersion is responsible for the turbid appearance of fresh VOO (Xenakis, Papadimitriou, & Sotiroudis, 2010). The popularity of this product is growing among some consumers and chefs, who consider the opalescent appearance signals greater freshness and quality (Zullo & Ciafardini, 2018). A colloid, homogeneous and non-crystalline, consist of large molecules or ultramicroscopic particles of one substance dispersed through a second substance. Freshly produced VOO has a cloudy appearance, since it contains variable amounts of micro-droplets of vegetation water and solid dispersed particles from olive fruit (Koidis, Triantafillou, & Boskou, 2008). Thus, to clarify, it is a complex colloid emulsion-sol.

While elaborating VOO, through main stages of grinding, malaxation and centrifugation, the transfer from the fruit mesocarp to the VOO of several minor components takes place (Gómez-Herrera, 2007). These minor parts are vegetation water and chemical species present in the olives, as well as species from reactions chemical or enzymatic, produced in the pulp from grinding (Gómez-Herrera, 2007).

Most modern mills base on separation by centrifugation, with some still using the classical hydraulic pressure method. Centrifugation mills employ two different alternative methods. One of them is the so-called three-phases. This employ some water added to the olive paste to ease extraction, separating the oil, the alpechin and the solid phase or pomace. Some of the last three-phase system versions use optionally a low addition or nothing of water. The other method, called two-phases, is more recent and does not use water, ensuring the beat up and separation by the vegetation water of the olives. This last provide a residue, the sludge or 'alperujo', a phase in which the vegetation water and the solid residues are together. Although the two methods perform similar, the antioxidant content of olive oil has been proven higher with the two-phases (Di Giovacchino, Sestini, & Di Vincenzo, 2002). This is mainly due to the washing effect produced by the added water in the three-phase method, which drastically reduces the content of polar compounds of olive oil, including a significant part of the phenolic compounds.

The matter suspended in the olive oil (referring to solids and moisture onwards) changes with the extraction method. With the press traditional extraction method, it can reach around 8% (Lercker, Frega, Bocci, & Servidio, 1994). Suspended solids in continuous extraction method in a specific study, both with two and three phases, were about 0.01% after the vertical centrifuge (Koidis & Boskou, 2006). In another later study, Koidis, Triantafillou, & Boskou (2008) described the suspended solids of an extra virgin olive oil got by the continuous extraction. They were small quantities of solid particles, around 12-460 ppm, with sizes in the range of 5 to 60 μm. This last study did not mention the method of the continuous extraction, two or three phases.

After a few weeks or months of storing VOO, the dispersion vanish and the opalescence disappears (Zullo & Ciafardini, 2018). Thus, we consider the fresh or young VOO as a perishable product. Fresh, opalescent or veiled VOO is a special type of olive oils. A suitable name for these products would be 'Integral' VOO. However, this term could lead to confusion with 'Integrated Production', widely spread in olive cultivation in Spain, the main producer, that is different (Cayuela, García, & Gutiérrez-Rosales, 2006). It is common to use market qualification as 'Gourmet', or 'Premium', referred to Extra VOO. These terms do not necessarily refer to fresh unfiltered Extra VOO. This last highlights by its content of micro-droplets of water and organic solutes in temporary dispersion, as well as by its perishable character. Most of fresh veiled VOO match to Extra class (EVOO), and usually only this is sold without filtering.

Nevertheless, we will refer it hereafter as VOO, since sometime it may not match to EVOO. It depends from subsequent analyses, reflecting the olive fruit quality.

The niche market kept by these olive oils so far, known as unfiltered VOO, matches mainly to the local demand of consumers, who know the product. The olive oil industry does not intend a large production of unfiltered VOO, since its demand is still minor. In addition, the global olive oil market needs an elastic adjustment overtime from supply to demand, which is not possible for this fresh olive oil. The consume of VOO is either fresh, this filtered or unfiltered, or stored in tanks to settle for weeks or months (Koidis, Triantafillou, & Boskou, 2008). A prolonged storage implies costs for the industry, thus bottling is frequent before the total sedimentation, preceded by pressure filtration through cotton filters (Guerrini & Parenti, 2016). Producers usually consider the best period from production to consumption is 12-18 months. Therefore, consumption of olive oil produced in a campaign is usually before the next season. In addition, the VOO of the previous season suffers a serious price decrease at the beginning of the campaign.

Some producers sell fresh virgin olive oils unfiltered, usually under the category Extra. This is the veiled or integral produce. Another part is packaged after filtering, without resting in tanks. This is frequent in small milling plants producing high quality olive oil (Guerrini & Parenti, 2016). However, most of virgin olive oil passes the phases of resting and storage. After that it is filtered under pressure with cotton filters, and the product is bottled, generally in packaging industries (Zullo & Ciafardini, 2018). Several marketing standard of olive oils compel filtrating. The last referred authors have stressed that filtration is one of the most controversial steps in producing olive oil. When the filtration and bottling is without a prior rest period, the olive oil may become cloudy in the bottle again, that is not desirable (Koidis, Triantafillou, & Boskou, 2008).

Colloid Stability of Fresh Virgin Olive Oil

The "veiled" VOO can keep its cloudy shape for several weeks or even months before the total deposition of the residue (Koidis, Triantafillou, & Boskou, 2008). Olive oil sensory analysis shows its sensory quality is better the shorter the time elapsed since its elaboration (Valli, Bendini, Popp, & Bongartz, 2014). However, VOO have a long shelf life thanks to its composition of triglycerides, in which oleic acid predominates, as well as its phenolic compounds and other antioxidants. The storage allows to gradually decant the vegetation water and other scattered matter, disappearing the turbidity. Thus, the sedimentation leads to separate two phases, the olive oil liquid phase and the sediments of water and solids previously forming a dispersion. For this purpose the product is left to stand in tanks of variable capacity, up to about 100 tons.

It seems logical to think the stability of the dispersion depends on the small size of the micro-droplets, which slows settling. This idea coincides with that previously reported by Guerrini & Parenti (2016). They highlighted the characteristics of the colloid's matter, affect the sedimentation time. However, fresh and unfiltered extra virgin olive oils are reported as metaestable emulsions or dispersions, involving emulsifying agents (Lercker, Frega, Bocci, & Servidio, 1994). About this, several authors have reported that compounds such as diacylglycerols, monoacylglycerols and phospholipids can act in freshly produced virgin olive oils as natural emulsifiers (Frega, Mozzon, & Lercker, 1999; Kiosseoglou & Kouzounas, 1993, Koidis & Boskou, 2006). These surfactants contributes to form an emulsion with the small amount of water present. This condition may persist for several months before becoming a separate phase or deposit. The deposit at the bottom of the industrial container, or at the consumer's bottle, is a brown residue of solids and some water. This deposit is unacceptable to consumers (Lercker, Frega, Bocci, & Servidio, 1994).

The diversity of the period the fresh olive oil preserves its veil in different products, is an important feature. The cases are different when this period is, *v.g.*, two weeks, or when it lasts two months. Zullo & Ciafardini (2018) referred the low physical stability of unfiltered VOO, as a key factor limiting its wider market distribution. Therefore, we suggest that 'colloidal stability' of VOO is an important new idea, that should not confuse with its oxidative stability. Nevertheless, close relations between both stabilities have been proven as forward shown. The time the colloidal dispersion last, will respond to the set of physiochemical features of the VOO. The measures for predicting or estimating the colloidal stability have not been defined, up to date. Besides, normally there is not a detailed checking on the vanish of the colloid state during storage, since there are few means to do it. There are at least two mechanisms whereby colloid stability is generally imparted, which are electrostatic stabilization and steric stabilization (Napper, 1970).

A recent study (Veneziani et al., 2018) has reported scanning electron microscopy (Cryo-SEM) analysis of veiled virgin olive oils, showing the presence of micro-dispersed water particles that did not contain apparent vegetable fragments. The study was focused on the compositional differences between veiled and filtered virgin olive oils during a simulated shelf life. In that study, portions of the veiled and filtered VOOs were frozen in liquid nitrogen and transferred to Cryo-SEM analysis.

A previous study reported the structure and dynamics of colloidal dispersions of veiled VOO, extracted by two-phase and three-phase systems, from one same olive fruit batch (Papadimitriou et al., 2013). The authors used confocal microscopy and the dispersive techniques "Small Angle Light Scattering Apparatus" (SALSA), "3D-Dynamic Light Scattering" (3D-DLS), "Classical Dynamic Light Scattering" (Green-DLS) and "Small Angle X-ray" Scattering "(SAXS). Both dynamic (3D-DLS) and static (SALSA) light scattering pointed out the presence of colloids in the micrometer region, while the SAXS experiments revealed the absence of colloids in the nanometer region. SALSA was the most suitable technique for characterizing samples of turbid VOO. The SALSA data showed a colloid diameter range between 1.5 and 14 μm, under the extraction conditions of the studied VOO. However the resolution limit of the method used was 15 μm, higher colloids being no detected. It should be noted that systems such as veiled olive oil have multiple dispersion. Besides, in systems where very large and very small particles are together, large particles scatter light much more than small ones, therefore small particles frequently are not detected. In the presence of particles of micrometric size and others in the nanometric range, a lot of small particles would be needed in the system for their contribution to be noticed (Chu & Liu, 2000). All this leads us to think that although in the study of Papadimitriou et al. (2013) nanometric particles were no detected, as the same authors warned, maybe these were present.

Confocal microscopy assessment did show with the two phases method, in the same referred study, a larger colloidal radius of the VOO sample from the three-phase method, with range 1-5 µm, respecting 1-2 µm. The SALSA measures showed the dispersed colloids underwent with time a significant decrease in their average radius from 7 to 1.2 µm in the two-phase sample, while the maximum size did not change. In the EVOO of three phases, the sizes followed the opposite trend. Both the average radius and the maximum size increased to 3 and 9.2 µm, respectively.

The authors (Papadimitriou et al., 2013) suggested that external water addition during the three-phase method, induced by coalescence the trend to larger colloid sizes. Assessment of EVOOs by confocal microscopy and SALSA coincided fully with that above. Added measurements after 6 months did not show essential changes. These results revealed that variable size of the colloids scattered in the EVOOs of three phases matches to water bags, relatively large, trapped inside the VOO mass.

A question that arises considering the previous concepts is the increase of colloids size may match in part to the volume increase of formations which can be named pseudo-vesicles. The different combinations of the minor surfactant ingredients present in fresh VOO can originate colloidal physical associations, such as reverse micelles and lamellar structures (Papadimitriou et al., 2013). Nevertheless, the hypothesis that part of the olive oil colloid may match to reverse micelles, vesicles, or pseudo-vesicles, is not explicit in the literature, to what we know. The amphiphilic molecules that can form these hypothetical structures in olive oil are proteins, sugar glycosides, glycoproteins, and to a lesser extent, phospholipids, according to contents reported in fresh olive oils (Lercker, Frega, Bocci, & Servidio, 1994; Koidis & Boscou, 2006). The free fatty acids are also amphiphilic, although their possible implication in this same role have not been considered up to now. They are responsible of the olive oil acidity. Thus, one hypothesis is the link of the free fatty acids to the referred structures may influence their own chemical activity, being expected to reduce them. This could be the case of their prooxidant effects, revealed by Frega, Mozzon, & Lercker (1999). Peptides and proteins have been traditionally considered as impurities of edible oils, but nowadays their presence is considered specially important, since it is related with oil stability (Hidalgo & Zamora, 2006). Although the VOO contents of these chemical species as a whole are quite low, their possible influence on the colloidal stability is also an interesting issue not studied. As well, their influence related to surfactant effects on freshness evolution of EVOO is poorly known.

To help clarify this issue, we have observed under microscopy some veiled olive oils. This analysis was made in the laboratory of the Unit of Microbiology of the Instituto de la Grasa (CSIC). For this purpose we used an upright optical microscope Eclipse Ni-E (Nikon, Japan). The technique used was differential interference contrast microscopy (DIC), coupled to a camera. An extra virgin unfiltered olive oil sample taken at the industry was observed. It was elaborated by the two-phases system and provided within a research project. The olive oil sample was taken at the decanter, once the elaboration process was finished. Its moisture content, determined by the official gravimetric method in the laboratories of Instituto de la Grasa (CSIC), was 0.34 %. The photographs showed micro-droplets of water, as well as numerous bulbous formations that appear to be colloidal 'water bags' (CWB), of varied sizes. A second olive oil sample was analyzed. It was a commercial unfiltered product from the two-phases system, labeled as virgin olive oil. Its measured moisture content was 0.28 %. Distilled and autoclaved water was added directly to 2 mL of unfiltered olive oil to obtain a total amount of 20% added water, then it was homogenized just before putting a drop on the slide for its microscopy observation. The original olive oil sample was kept at 4 ºC for about 20 days, during which 8 new olive oil samples with 20% added water were prepared. Both the samples of the original olive oil and the samples of olive oil homogenized with the added water provided separately very similar images in the sequential observations. We have observed moving material within these CWB. The physic influence of the internal electrostatic charges may explain the movement observed in the particles contained therein. This may be the case of a CWB from a VOO sample without any water addition, examined inside during the first 23 seconds with 40X objective, and then outside with 10X objective (Video 1).

Compared to VOO samples homogenized with 20% water, CWB appeared different (Figures 1-4). In Figure 3 the irregular contour of a CWB can be clearly appreciated, as well as what appear to be small drops of water adhered externally. When adding water, perfect spheres appears to match with confined water inside the triglyceride liquid (Figure 2 and Figure 4). The existing literature have not indicated these observations.

Our hypothesis is that amphiphilic molecules present in the veiled VOO may locate at the interface of the CWB, with their hydrophilic and lipophilic parts in the corresponding medium. This does not occur in inclusions or pockets of added water. Said molecules likely do not get to form a true vesicle, but could form a pseudo-layer that protects to some extent the inner content of the aqueous phase.

Colloid and Dispersed Matter

The matter dispersed in fresh olive oils match to micro-droplets of vegetation water and solid particles from the fruit pulp. In a detailed description of the physiochemical parts of olive pastes, Gómez-Herrera (2007) distinguished as different parts VOO, vegetation water, bone fragments and pulp. The pseudo-colloidal pulp of the olive paste has a high water content. It consists on a set of many fragments of minute size from the mesocarp's crushing, in a colloidal state similar to that of a pseudo-micro-gel.

Both the appearance and the color of the olive oil depend on the turbidity produced by the dispersed matter (Gordillo, Ciaccheri, Mignani, Gonzalez-Miret, & Heredia, 2011). Lercker, Frega, Bocci, & Servidio (1994) reported nitrogen content 0.6% on wet matter in the deposit waste, most likely in the form of protein substances. We understand that could translate, considering a protein factor 5.75, into contents close to 0.1% protein. Koidis & Boskou (2006) researched in a later study the content of proteins and phospholipids of veiled virgin olive oils, reporting contents of protein levels below 2.5 mg/kg and 21–124 mg/kg of phospholipids.

It is interesting to highlight two facts that seem proven. The two-phase method provides a highest content of antioxidant compounds (Di Giovacchino, Sestini, & Di Vincenzo, 2002). In this, the colloidal size is smaller (Papadimitriou et al., 2013). Thus, can deduce that VOO quality preserves better from oxidation when smaller the colloid size. The reported increase of colloids size matches to relatively large water bags. Thus, it is logical think that phenolic compounds can no exert an antioxidant role while trapped inside it. However, there is no information on this question. Meanwhile, the microdroplets of water suspended directly in the triglyceride mass, likely content also amphiphilic phenolics. Considering the information above, the role of the different components of fresh veiled EVOOs on the colloidal stability during storage, is far from being well known. Specific measures to characterize the decantation of water micro-droplets in the tanks of olive oil, in terms of dynamic physics, are not reported in literature. Perhaps it would be possible considering separately how evolves the solid particles and water micro-droplets, scattered in the triglyceride matrix.

Moisture and Phenolics of Virgin Olive Oil

Seemingly, the main cause of turbidity in unfiltered virgin olive oils is the content of water droplets (Lercker, Frega, Bocci, & Servidio, 1994; Koidis & Boskou, 2006). Other authors have reported that said micro-droplets have sizes between 1 and 5 µm (Koidis, Triantafillou, & Boskou, 2008). Water content in fresh olive oil is variable, with an average value of about 0.5% by weight (Papadimitriou et al., 2013). According to our own data, in certain freshly extracted virgin olive oils water can exceed 1%, reaching up to about 1.5% (data not reported). A cause that significantly influences the water content of olive oils is the conditions of the centrifugation treatment (Guerrini & Parenti, 2016).

The vegetation water present in the VOO may contains enzymes and, in particular, lipase. It is able to hydrolyse triglycerides to release free fatty acids, that increasing the acidity of VOO(Di Giovacchino, 2013). In fact, the enzyme acylhydrolase, together with lipoxygenase, and fatty acid hydroperoxide lyase, were found in cell-free extract of olive fruit (Olías, Pérez, Ríos, & Sanz, 1993). In damaged olives, before processing and during transformation, the lipases may be responsible for the high olive oil acidity due to the hydrolysis of the triglycerides. High acidity exerts a great influence on olive oil stability (Frega, Mozzon, & Lercker, 1999; Ciafardini, Zullo & Iride, 2006), and a prooxidant effect of free fatty acid content of olive oils has been highlighted (Frega, Mozzon, & Lercker, 1999). Moreover, lipase from yeasts has been reported also in olive oil (Ciafardini, G., Zullo, B. A., & Iride, A., 2006; Ciafardini & Zullo, 2018).

In the other hand, water is rich in water-soluble phenols, and these provides a strong antioxidant effect (Lercker, Frega, Bocci, & Servidio, 1994; Ambrosone, Angelico, Cinelli, Di Lorenzo, & Ceglie, 2002). Thus, the presence of water could lessen indirectly the rate of oxidation, improving VOO stability. It is interesting to remember there is a large literature, including multiple reviews, on the antioxidant features of the olive oil phenolics. Both about their contribution to the stability of the product, as well as on the properties making them healthy (Tsimidou, 1998). Can highlight that generalization of the use of hammer mills has facilitated a greater permanence of phenolic compounds than when grinding in traditional stone mills (Caponio, Gomes, Summo, & Pasqualone, 2003). Therefore, the olive oils held have greater antioxidant capacity (Di Giovacchino, Sestini, & Di Vincenzo, 2002). The main phenolic compounds of virgin olive oil are aglycones of oleuropein and its derivatives, among them the simple phenols tyrosol and hydroxytyrosol. These compounds are markedly polar and water soluble (Galanakis, Goulas, Tsakona, Manganaris, & Gekas, 2013), so their presence in the olive oil depends on their water content (Lercker, Frega, Bocci, & Servidio, 1994; Ambrosone, Angelico, Cinelli, Di Lorenzo, & Ceglie, 2002; Di Giovacchino, Sestini, & Di Vincenzo, 2002), which is drastically reduced by filtration treatments.

Where the olive fruit water content locates is not clear, to our knowledge, despite reports on the microscopic analysis of the olive mesocarp (Rallo & Rapoport, 2001). The cells are almost all occupied by their central vacuoles, which are hydrophobic since they are full of triglycerides. Thus, the water must necessarily be in the intercellular spaces. It is logical to think the amphiphilic compounds of the olive oil may locate in the interface of the external limits of the vacuoles. During olives milling, the parenchyma tissue forming the mesocarp transforms irregularly, resulting in the different parts that can distinguish in the microscopic study of the olive pastes (Gómez-Herrera, 2007). The milling release both the VOO from the vacuoles of the parenchyma cells, and the vegetation water. The olive oil coalesce during the beat up. In this stage, the vegetation water forms a continuous phase in contact with other parts of the paste (Gómez-Herrera, 2007). Until now, the stage of the elaboration of VOO in which the water microscopic droplets appear, is not described. It is reasonable to think the centrifugation treatment matches to its formation and dispersion in the lipid, making up the moisture present in the fresh VOOs. Since the VOO phenolic compounds are quite water soluble, their presence in the olive oil depends on their moisture content. The findings of a recent research (Veneziani et al., 2018) matches to these ideas. The specific settings of the vertical centrifuge used to treat four industrial VOO samples extracted in different Mediterranean areas, determined the 'veiling' stabilization and reduced the formation of deposits at the bottom of the oil bottles. As well, in the same study, a higher phenolic concentration was detected at the end of storage period in the veiled olive oils compared to filtered samples.

Bakhouche et al. (2014[a]) assessed in industrial filtration tests, before olive oil storing, how filtration affects to the moisture content and phenolic compounds. The study highlighted that filtration drastically reduced humidity, decreasing phenolic alcohols and flavones, which are phenols compounds water soluble. Among them are important hydroxytyrosol and tyrosol (Capasso, Evidente, & Scognamiglio, 1992; Bakhouche at al., 2014 [a]). These last authors reported also that secoiridoids increase, while lignans were the least affected group. However, the same group (Bakhouche et al., 2014[b]) relativized the increase of secoiridoids in a later work, in which using an internal oleuropein standard they inferred said increase was not. The most notable conclusions of Bakhouche et al. (2014 [a]) were that filtration can increase the useful life of EVOOs by reducing their moisture content. But it sacrifices part of the phenolic compounds, which can affect their oxidative stability, and its nutritional quality. The authors settled that to preserve the olive oil quality, it is important to consider both the loss of moisture and the content of antioxidants during filtration. This suggests, we think, that it is possible redefine a best moisture content for fresh olive oils, despite the International Olive Council suggested an generic upper limit of 0.2% (IOOC, 2009). In this case, may consider the possibility of centrifugation treatments to regulate water content.

It is also worth remembering that scientific literature has reported the moisture present in olive oil could links to fermentation during elaboration (Ciafardini, Zullo, Cioccia, & Iride, 2006; Cayuela, Gómez-Coca, Moreda, & Pérez-Camino 2015; Zullo & Ciafardini, 2018; Ciafardini & Zullo, 2018). Different types of living microorganisms have been found in fresh VOO, such as bacteria, yeasts and fungi (Ciafardini, Zullo, Cioccia, & Iride, 2006; Koidis, Triantafillou, & Boskou, 2008; Mari, Guerrini, Granchi, & Vincenzini, 2016). The concentrations of microorganisms are low, like 3 log cfu/mL

(Koidis, Triantafillou, & Boskou, 2008). These microorganisms come from the epiphytic microflora of the fruit. Their enzymatic activities do not seem to affect the quality of the final product (Koidis, Triantafillou, & Boskou, 2008). However, the influence of microorganisms on the olive oil quality evolution depending on its moisture content and conservation procedure, is far to be well-known (Cayuela, Gómez-Coca, Moreda, & Pérez-Camino, 2015). Greater microbial activity and higher quality decrease has been reported in VOO with lower contents of non-polar phenolics (Zullo & Ciafardini, 2018). In a previous study, it was proven that olive fruit can receive microbial contamination in the washing hoppers, that was related to the presence of fermentative defects in the product (Vichi, Romero, Tous, & Caixach, 2011). It is also reported that in a few hours after elaborating olive oil, yeasts such as Candida and Saccharomyces can develop colonies (Ciafardini, Zullo, Cioccia, & Iride, 2006). The microflora thus set up can survive throughout the storage period, and could damage the sensory characteristics of the product (Guerrini & Parenti, 2016). Besides, Di Giovacchino (2013) reported the sediments on the bottom of the VOO containers can ferment under certain temperature conditions. These sediments contains sugars, proteins and enzymes. Its fermentation produces some substances, *v.g.* short-chain fatty acids, that give a typical defect of muddy sediment or putrid. In unfiltered olive oils, this could perhaps happen sometimes even after packaging, depending on the consumption date.

Volatile compounds

The sensory characteristics of olive oils depends largely on its volatile compounds (Jacini, 1976). Thus, it is interesting studying the differences in these compounds between fresh colloidal olive oils and filtered VOO. Filtration using a polypropylene filter bag and argon or nitrogen gas flows as filter aids did not decrease the intensity of the main positive sensory attributes, according to Lozano-Sánchez et al. (2012). Brkić Bubola, Koprivnjak, & Sladonja (2012) studied the change induced by filtration to the VOO volatiles profile, using hydrophilic cotton, pointing to unequal filtration impact on different monovarietal olive oils. Buža cv. showed a slight increase in total alcohols, while a significant decrease of total alcohols and slight changes in total aldehydes in Črna cv. were observed. No significant influence on the sensory scores of olive oils, but some slightly higher intensities of sensory characteristics apple and grass, were noticed.

Sacchi, Caporaso, Paduano, & Genovese (2015) reported that majority of 38 volatile compounds analyzed by the SPME-GC/MS technique did not change significantly after industrial filtration, while some of them increased their initial concentration, and others significantly decreased. Some volatiles which increased after filtration in the referred study were 6-Methyl-5-hepten-2-one and heptanol, while 2-methylbutanal increased up to twofold. Besides, t-2,4-hexadienal, t-2-hexen-1-ol, and c-2-penten-1-ol significantly decreased.

The possibility of removing by filtration unpleasant olive oil flavors has been also investigated. This was the case of a study focused on some compounds responsible for the specific unpleasant flavor 'eucalyptus' (Bottino, Capannelli, Mattei, Rovellini, & Zunin, 2008). The referred filtration treatments were made by cross-flow microfiltration (MF) and ultrafiltration (UF) with different commercial membranes. The study concluded that UF Carbosep membrane was the most suitable for softening the olive oil organoleptic features, regarding the unfiltered olive oils. Moreover, filtration has been recommended to circumvent the presence of short-chain alcohols, mainly ethanol and methanol, once the olive oil has been extracted (Gómez-Coca, Fernandes, Pérez-Camino, & Moreda, 2016). In this last study the relationship between the olive oil content of ethanol and methanol and the formation of alkyl esters (FAAE), has been shown. This formation occurs in the presence of free acidity. As well, the same study has pointed out the relationship between FAAE and the main VOO fermentative defect. We have not found more information regarding the influence of filtration on olive oil volatiles. Studying the differences in the formation of volatile compounds between fresh colloidal olive oils and the same products after filtration is, therefore, a particularly interesting topic.

Oxidative Stability, Moisture and Dispersed Matter

While stored, VOO undergoes much changes in its chemical and sensory characteristics, since it is prone to various oxidative degradation, such as enzymatic oxidation, photoxidation and autooxidation. The enzymatic oxidation was reported by

Georgalaki, Sotiroudis, & Xenakis (1998). As well, relationships of virgin olive oil endogenous oxidoreductases with phenolic and volatile compounds during the mechanical extraction process of olive oil, was revealed by Servili, Baldioli, Begliomi, Selvaggi, & Montedoro (2000). Bendini, Cerretani, Salvador, Fregapane, & Lercker (2009) reviewed in detail the causes involved in the auto-oxidation, which is the main cause of quality decay of olive oil during the commercial life. Many solutions have been proposed to preserve its quality, and to extend its useful life (Guerrini & Parenti, 2016). The control of the causes involved in oxidation is basic for conserving olive oil quality during storage and shelf life (Bendini, Cerretani, Salvador, Fregapane, & Lercker, 2009), a key issue for the industry. Nevertheless, only some studies on olive oil quality conservation considers the colloidal features of unfiltered VOO.

Lercker, Frega, Bocci, & Servidio (1994) reported in a short communication their research on the effects on shelf life of dispersed matter of Extra VOO. Their results proved the oxidative stability was higher in unfiltered olive oil samples, press extracted. Therefore, they suggested the material in dispersion, roughly 8% of olive oil in that study, was responsible for its better stability. The authors analyzed the influence of the scattered matter on the acidity increase. Olive oil acidity comes from free fatty acids, produced by enzymatic hydrolysis. They proved the free fatty acids links to the residue, precipitating with this. The authors related the precipitation of the dispersed particles to opposite electrical charges attraction. A no negligible amount of nitrogen was found in the deposited waste, 0.6% on wet matter, most likely in the form of protein substances. The conclusion of the research was the suspended particles making up the "veil" of fresh VOO, played a stabilizing role in its useful life. They acts as antioxidants, and as a buffer against the increase of the acidity, according the referred study. Thus, do not filtering was reported as a beneficial practice.

However, several other studies agree that acidity in stored unfiltered olive oils increases respecting to the same filtrate, despite the oxidative stability remains slightly better (Tsimidou, Georgiou, Koidis, & Boskou, 2005; Fregapane, Lavelli, Leon, Kapuralin, & Salvador, 2006; Stefanoudaki, Williams, & Harwood, 2010). In this same sense, it is reported the enzymes remaining after olive oil filtration could be uninhibited to catalyze the oxidation of polyunsaturated fatty acids. This may be because some antioxidants are among the minor ingredients removed. They locates close the enzyme as amphiphilic molecules previously to filtration, preventing their catalytic action (Georgalaki, 1999). However, other possible hypothesis is the enzymes may be joint before filtration to the CWB reported in the present study, where its activity could be reduced or inhibited. Frega, Mozzon, & Lercker (1999), studying the effects of free fatty acids on the oxidative stability of vegetable oil, showed that dispersed particles play a double stabilizing effect on both oxidative and hydrolytic degradation. From their results, the authors suggested that avoid filtrating extra virgin olive oil is desirable to prolong its shelf life. As well, several authors (Lercker, Frega, Bocci, & Servidio, 1994; Koidis, Triantafillou, & Boskou, 2008) proved the influence of natural emulsifiers present in olive oils on its oxidative stability. Besides, Ambrosone, Angelico, Cinelli, Di Lorenzo, & Ceglie (2002) filtered out the solid particles content of fresh olive oils, adding water under controlled conditions. Then, they oversaw the olive oil oxidation, reporting that dispersed water reduces the speed of its oxidation. However, reasons why water microdroplets itself may reduce the oxidation rate of VOO, independently from phenolics, were not suggested. These reports agree on benefits from do not filtering.

Sotiroudis, Sotiroudis, Varkas, & Xenakis (2005) reported a study of the oxidative stability VOO emulsions with water, using emulsifying agents from olive oil, and suggesting the presence of stabilizing structural olive oil proteins.

Tsimidou, Georgiou, Koidis, & Boskou, (2005) investigated the loss of oxidative stability during the storage of unfiltered veiled virgin olive oils. They carried out a nine-month storage trial at room temperature in the dark. The study was carried out on VOO samples filtered in the laboratory using a common filter paper, according to the authors. They reported a lower stability of the filtered veiled VOO, that assigned to the fall of the total polyphenol content. They signaled a gradual loss of oxidative stability in filtered olive oils after nine months of storage at room temperature in the dark. This fact agrees with a greater increase of the peroxide values in the filtered samples, as well as with the decrease of polar phenols content. The authors suggested the individual phenol compounds released by hydrolysis in unfiltered olive oils may provide greater oxidative stability (Tsimidou, Georgiou, Koidis, & Boskou, 2005). These conclusions signaled is better not filtering. The studies commented in the previous paragraphs matches in recognizing the scattered matter of fresh VOO exerts favorable effects, slowing down their oxidation and improving their stability. Fregapane, Lavelli, Leon, Kapuralin, & Salvador (2006) have examined the effect of dehydrating and filtrating on the stability and quality of VOO during storage, both at 25ºC and at 40ºC. They compared the evolution olive oils unfiltered, filtered and dehydrated from several monovarietal VOO. The results showed filtrating and dehydrating decreased the hydrolysis rate of the triacylglycerol mass. This effect was proportional to the increase in temperature and acidity. These treatments delayed the beginning of the rancid defect. The hydrolysis of secoiridoid derivatives forming simple phenols was higher in unfiltered olive oils. The authors inferred that filtrating and especially dehydrating could help prolong the shelf life of high-quality olive oils. The effect was larger for the less stable olive oils, such as those from the Arbequina and Colombaia varieties. These results contradict partially that of Ambrosone, Angelico, Cinelli, Di Lorenzo, & Ceglie (2002), about that scattered water lessens the speed of its oxidation, and disagree on benefits from do not filtering VOO.

Stefanoudaki, Williams, & Harwood, (2010) conducted a study on the VOO quality evolution under different storage conditions. The last authors signaled their results did not coincide at one point with those of Tsimidou, Georgiou, Koidis, & Boskou (2005). The mismatching was on a greater phenolic loss and lower oxidative stability of filtered olive oils respecting that of unfiltered olive oils. They found a greater increase in acidity in unfiltered oils at 8 and 10 months in a storage trial of both types of oils. Besides, the differences in the values of peroxides, dienes and conjugated trienes between both types of olive oils, were not significant considering the overall results of the trial. Thus, this disagree on benefits from do not filtering VOO.

The influence of new filtration systems on the quality of extra virgin olive oil has been subject of study. Lozano-Sánchez et al. (2012) reported that filtration processes using a polypropylene filter bag and argon or nitrogen gas flows as filter aids. The authors highlighted the filtration using inert gases did not decrease the intensity of the main positive sensory attributes, assessed by sensorial analysis. They confirmed the higher water content of unfiltered olive oils. As well, they found a slight tendency of oxidative stability to decrease after filtration. Filtering using argon gas flow and a filter bag were the most effective for reducing the water content. These last olive oils had the lowest polar-phase contents. This effect was more pronounced when the argon gas and filter bag were used as filter aids than when using nitrogen gas (Lozano-Sánchez et al., 2012). That effect could be related to the lower water content in the first filtration system, which consequently lowers the concentration of polar phenols, and therefore the antioxidants contents. Moreover, it was shown that polar-phenolic compounds oriented in the water-in-oil emulsion interface are more protective against oxidation (Bendini et al., 2007).

Besides, Guerrini & Parenti (2016) highlighted that improving the stability of virgin olive oils, needs storing it without high amounts of suspended solids and water. They signaled for this purpose is suitable removing water from fresh olive oil. This opinion pours into a monographic chapter dedicated to review vertical centrifugation and filtration. This chapter considered the negative effects of these treatments on the evolution of olive oil quality during storage. As well, the authors reviewed the possible technical solutions proposed so far. Their discussion of the literature did not deepen on the links between the disperse matter in fresh olive oils and their stability.

In a recent study, Brkić Bubola, Luki, Mofardin, Butumovi, & Koprivnjak (2017) compared quality changes of virgin olive oils clarified by filtration at an industrial scale and those clarified by natural sedimentation and decantation. Natural sedimentation after 6 months provided higher delay of oxidative deterioration, while filtration provided a more stable sensory profile.

Zullo & Ciafardini (2018) have researched a new storage to pack fresh virgin olive oil avoiding rapid sedimentation, producing long-lived veiled VOO. The proposed method bases on turning the oil containers every 20 days, preventing sedimentation of the suspended materials. Also, the referred authors studied the changes in the physiochemical and microbiological features of veiled virgin olive oil during storage in the dark. They performed chemical and microbiological analyses on three short-lived veiled virgin olive oils (3-5 months) and three long-lived virgin olive oils (1-year), which contained different concentrations of polar phenolic compounds. The study found the best conservation in both types of veiled VOO when the content of polar phenolics was higher than 130 mg / kg (eq to coffee acid). The solid particles present in the short-lived veiled virgin oil settled on the bottom of the containers (98%), producing sediments, during storage for 6 months. On the contrary in the long-lasting veiled olive oil, got by the tumbling method referred, only 2-6% of the solid particles settled.

It might seem there is a certain contradiction about the benefit or harm from the sediments in olive oil storage. Janakat, Al-Nabulsi, Hammad, & Holley (2015) studied olive oil from different origins during 12 months. The authors compared olive oil normal, olive oil extracted from sediment, and product taken from the bottom of the deposit, nearby the sediment. The authors deduced from their results the sediment in the deposit during the storage of olive oil stabilized its quality. However, Di Giovacchino (2013) recommended separate VOO from the sediment as quickly as possible, either by pouring or by filtering it through hydrophilic materials. Although the question is controversial, it seems there is more consensus on separating VOO from the sediment as quickly as possible is the best to avoid the 'muddy sediment' or 'borras' defects.

These seeming contradictions on benefits from VOO dispersed matter involves, in our opinion, the physiochemical state of VOO. The opalescent metastable dispersion the fresh VOO colloid is, splits with long time into two phases. One phase is the liquid VOO, with a high degree of transparency, the other one is a residual sediment. In this evolution EVOO loses its freshness, in the sense of losing the 'integral' product feature. Clarifying this seeming contradiction can help to improve the overall figures of product quality. Nearly one half of olive oil production matches to EVOO, and other half with sensory defects to some extent (Cayuela, Gómez-Coca, Moreda, & Pérez-Camino, 2015).

The results of the studies above detailed, suggest several ideas. During the time the fresh VOO remains as a colloid, it seems its quality features remain significantly closer to the original. As well, these results indicate that after disappearing the colloidal dispersion of micro-water droplets and organic aggregates, begins decaying its quality.

Therefore, it is of great interest to improve the knowledge on the colloidal stability of the integral VOO, both in the storage tanks and commercial containers.

Future Trends

The benefits from the dispersed matter of fresh veiled VOO may involve their physiochemical state. Fresh VOO are not generally managed as perishable products. Thus, it is of great interest to improve the knowledge on the colloidal stability of veiled VOO during their conservation. As well, it is interesting to know its evolution in fresh packaged EVOOs. In this sense, it is important to tune up methods for estimating the colloidal stability of veiled VOO.

In short, it would be interesting to better know the evolution of the colloidal physiochemical state of VOO during storage and market life. It is desirable to know the physical colloidal features of the VOO, especially into the signs of the time when the colloidal dispersion losses its stability. This could provide objective data on when the integral VOO vanish. They can also help to control the best moment of packaging, with or without filtration.

A better knowledge on the specific quality characteristics of fresh VOO due to its colloidal physiochemical state, will be useful for deepening the improvement of this productive sector. Can be highlighted the interest on the relationships between the stabilities colloidal and oxidative of veiled VOO.

Acknowledgement

The authors thank the Spanish National Council of Scientific Research (CSIC) for the means it provides at the disposal of its personnel. This review work has not received specific funding. We acknowledge also to Dra. Olga López Serrano, responsible of the research group ‘Lipid and Interphases Biophysics’, of the Institute of Advanced Chemistry of Catalonia (CSIC), for her suggestions and her kindness to collaborate on apossible research project. We are also grateful to S. A. T. Santa Teresa for providing the unfiltered olive oil sample to which one of the microscopy observations refers, within the research project Feder Innterconecta EXP 00091182 / ITC-20161186, ‘Olivesense’.

References

1. Ambrosone, L., Angelico, R., Cinelli, G., Di Lorenzo, V., & Ceglie, A. (2002). The Role of Water in the Oxidation Process of Extra Virgin Olive Oils. *Journal of the American Oil Chemists' Society, 79*, 577-581.

2. Bakhouche, A., Lozano-Sánchez, J., Ballus, C. A., Martínez-García, M., González Velasco, M., & Olavarría-Govantes, A. (2014[a]). Monitoring the moisture reduction and status of bioactive compounds in extra-virgin olive oil over the industrial filtration process. *Food Control, 40,* 292-299.

3. Bakhouche, A., Lozano-Sánchez, J., Ballus, C. A., Bendini, A., Gallina-Toschi, T.,Fernández-Gutiérrez, F., Segura-Carretero, A. (2014[b]). A new extraction approach tocorrect the effect of apparent increase in the secoiridoid content after filtration of virgin olive oil. *Talanta 127,* 18–25.

4. Bendini, A., Cerretani, L., Salvador, M. D., Fregapane, G., & Lercker, G. (2009). Stability of the sensory quality of virgin olive oil during storage: An overview. *Italian Journal of Food Science, 21*(4), 389-406.

5. Bendini, A., Cerretani, L., Carrasco-Pancorbo, A., Gomez-Caravaca, A. M., Segura-Carretero, A., Fernandez-Gutierrez, A., Lercker, G. (2007). Phenolic molecules in virgin olive oils: A survey of their sensory properties, health effects, antioxidant activity and analyticalmethods. An overview of the last decade. *Molecules, 12,* 1679−1719.

6. Bottino, A., Capannelli, G., Mattei, A., Rovellini, P., & Zunin, P. (2008). Effect of membrane filtration on the flavor of virgin olive oil. *European Journal of Lipid Science and Technology, 110,* 1109–1115.

7. Brkić Bubola, K., Koprivnjak, O., Sladonja, B., (2012). Influence of filtration on volatile compounds and sensory profile of virgin olive oils. Food Chemistry. 2012, 132, 98–103.

8. Brkić Bubola, K., Luki, M., Mofardin, I., Butumovi, A., Koprivnjak, O. (2017). Filtered vs. naturally sedimented and decanted virgin olive oil during storage: Effect on quality and composition. *LWT - Food Science and Technology, 84,* 370-377.

9. Capasso, R., Evidente, A., & Scognamiglio, F. (1992). A Simple Thin Layer Chromatographic Method to Detect the Main Polyphenols Occurring in Olive Oil Vegetation Waters. *Phytochemical Analysis, 3*(6), 270-275.

10. Caponio, F., Gomes, T., Summo, C., & Pasqualone, A. (2003). Influence of the type of olive-crusher used on the quality of extra virgin olive oils. *European Journal of Lipid Science & Technology, 105,* 201–206.

11. Cayuela, J. A., García, J. M., & Gutiérrez-Rosales, F. (2006). Influencia de la producción integrada del olivar sobre la calidad del aceite de oliva virgen. *Grasas y Aceites, 57*(4), 422-428.

12. Cayuela, J. A., Gómez-Coca, R. B., Moreda, W. & Pérez-Camino, M. C. (2015). Sensory defects of virgin olive oil from a microbiological perspective. *Trends in Food Science & Technology, 43,* 227-235.

13. Chu, B., Liu, T. (2000). Characterization of nanoparticles by scattering techniques. *Journal of Nanoparticle Research 2,* 29–41.

14. Ciafardini, G., Zullo, B. A., Cioccia, G., & Iride, A. (2006). Lipolytic activity of Williopsis californica and Saccharomyces cerevisiae in extra virgin olive oil. *International Journal of Food Microbiology, 107*(1), 27-32.

15. Ciafardini, G., Zullo, B. A., Iride, A. (2006). Lipase production by yeasts from extra virgin olive oil. *Food Microbiology, 23*(1), 60-67.

16. Ciafardini, G., Zullo, B. A. (2018). Virgin olive oil yeasts: A review. *Food Microbiology, 70*, 245-253.

17. Di Giovacchino, L., Sestini, S., & Di Vincenzo, D. (2002). Influence of olive processing on virgin olive oil quality. *European Journal of Lipid Science & Technology, 104,* 587.

18. Di Giovacchino, L. (2013). Technological aspects. In R. Aparicio & J. Harwood (Eds.), *Handbook of olive oil* (pp. 57-94). London: Springer.

19. Frega, N., Mozzon, M., & Lercker, G. (1999). Effects of free fatty acids on oxidative stability of vegetable oil. *Journal of the American Oil Chemists' Society, 76,* 325.

20. Fregapane, G., Lavelli, V., Leon, S., Kapuralin, J., & Salvador, M. D. (2006). Effect of filtration on virgin olive oil stability during storage. *European Journal of Lipid Science & Technology, 108,* 134-142.

21. Galanakis, C. M., Goulas, V., Tsakona, S., Manganaris, G. A., & Gekas, V. (2013). A Knowledge Base for The Recovery of Natural Phenols with Different Solvents, *International Journal of Food Properties, 16*(2), 382-396.

22. Georgalaki, M. D. (1999). *Enzymatic activities in oils and olive oil.* Agricultural University of Athens. (PhD thesis).

23. Georgalaki, M. D., Sotiroudis, T. G., Xenakis, A. (1998). The presence of oxidizing enzyme act in virgin olive oil. *Journal of the American Oil Chemists Society, 75*(2) 155-159.24. Gómez-Coca, R. B., Fernandes, G. D., Pérez-Camino, M. C. Moreda, W. (2016). Fatty acid ethyl esters (FAEE) in extra virgin olive oil: A case study of a quality parameter. *LWT - Food Science and Technology 66,* 378-383.

25. Gómez- Herrera, C. (2007). Matter transfer during virgin olive oil elaboration. *Grasas y Aceites, 58*(2), 194-205.

26. Gordillo, B., Ciaccheri, L., Mignani, A. G., Gonzalez-Miret, M. L., & Heredia, F. J. (2011). Inluence of turbidity grade on color and appearance of virgin olive oil. *Journal of the American Oil Chemists' Society, 88*(9), 1317-1327.

27. Guerrini, L., & Parenti, A. (2016). Stabilization of Extra-Virgin Olive Oil. In D. Boskou (Ed.), *Products from Olive Tree* (pp. 55-65). http://dx.doi.org/10.5772/64401. London: IntechOpen.

28. Hidalgo F. J., & Zamora R. (2006). Peptides and proteins in edible oils: stability, allergenicity, and new processing trends. *Trends Food Science & Technology, 17,* 56–63.

29. International Olive Oil Council. (2009). Trade Standard applying to Olive Oil and Olive Pomace Oils. COI/T.15/NC No2.

30. Jacini, H. (1976). Organoleptic characteristics of olive oil. *Grasas y Aceites, 27*(4), 255-258.31. Janakat, S., Al-Nabulsi, A., Hammad, F., & Holley, R. (2015). Effect of amurca on olive oil quality during storage. *Journal of Food Science and Technology, 52,* 1754.

32. Kiosseoglou, V., & Kouzounas, P. (1993). The role of diglycerides, monoglycerides, and free fatty acids in olive oil minor surface-active lipid interaction with proteins at oil-water interfaces. *Journal of Dispersion Science and Technology, 14,* 527-539.

33. Koidis, A., & Boskou, D. (2006). The contents of proteins and phospholipids in cloudy (veiled) virgin olive oils. *European Journal of Lipid Science & Technology, 108,* 323-328.

34. Koidis, A., Triantafillou, E., & Boskou, D. (2008). Endogenous microflora in turbid virgin olive oils and the physicochemical characteristics of these oils. *European Journal of Lipid Science & Technology,110,* 164–171.

35. Lercker, G., Frega, N., Bocci, F., & Servidio, G. (1994). “Veiled” extra-virgin olive oils: Dispersion response related to oil quality. *Journal of the American Oil Chemists' Society, 71,* 657-658.

36. Lozano-Sánchez, J., Cerretani, L., Bendini, A., Gallina-Toschi, T., Segura-Carretero, A., Fernández-Gutiérrez, A. (2012). New Filtration Systems for Extra-Virgin Olive Oil:

Effect on Antioxidant Compounds, Oxidative Stability, and Physicochemical and Sensory Properties. *Journal of Agricultural and Food Chemistry, 60,* 3754−3762.

37. Mari, E., Guerrini, S., Granchi, L., & Vincenzini, M. (2016). *World Journal of Microbiology and Biotechnology, 32,* 93.

38. Napper, D. H. (1970). Colloid Stability. *Industrial & Engineering Chemistry Product Research and Development, 9*(4), 467-477.

39. Olías, J. M., Pérez, A. G., Ríos, J. L., Sanz, L. C. (1993). Aroma of Virgin Olive Oil: Biogenesis of the "Green" Odor Notes. *Journal of Agricultural and Food Chemistry 41*, 2368-2373.

40. Papadimitriou, V., Dulle, M., Wachter, W., Sotiroudis, T. G., Glater, O., & Xenakis, A. (2013). Structure and dynamics of veiled virgin olive oil: inluence of production conditions and relation to its antioxidant capacity. *Food Biophysics, 8*(2), 112-121.

41. Rallo, L., & Rapoport, H. F. (2001). Early growth and development of the olive fruit mesocarp. Journal of Horticultural Science & Biotechnology, 76(4), 408-412.42. Sacchi, R., Caporaso, N., Paduano, A., & Genovese, A. (2015). Industrial-scale filtration affects volatile compounds in extra virgin olive oil cv. Ravece. *European Journal of Lipid Science and Technology, 117,* 2007–2014.

43. Servili, M., Baldioli, M., Begliomi, A. L., Selvaggi, R., Montedoro, G. (2000). The phenolic and volatile compounds of virgin olive oil: Relationships with the endogenous oxidoreductases during the mechanical oil extraction process. *Flavour and Fragance Chemistry, 46*, 163-173.

44. Sotiroudis, T. G., Sotiroudis G. T., Varkas N., Xenakis A. (2005). The role of endogenous amphiphiles on the stability of virgin olive oil-in water emulsions. *Journal of the American Oil Chemists' Society, 82*, 415–420.

45. Stefanoudaki, E., Williams, M., & Harwood, J. (2010). Changes in virgin olive oil characteristics during different storage conditions. *European Journal of Lipid Science &. Technology, 112,* 906–914.

46. Tsimidou, M. (1998). Polyphenols and quality of virgin olive oil in retrospect. *Italian Journal of Food Science, 10*(2), 99-116.

47. Tsimidou, M. Z., Georgiou, A., Koidis, A., & Boskou, D. (2005). Loss of stability of ‘‘veiled’’ (cloudy) virgin olive oils in storage. *Food Chemistry, 93,* 377–383.

48. Vichi, S., Romero, A., Tous, J., & Caixach, J. (2011). The Activity of Healthy Olive Microbiota during Virgin Olive Oil Extraction Influences Oil Chemical Composition. *Journal of Agricultural and Food Chemistry, 59,* 4705–4714.

49. Valli E., Bendini A., Popp M., & Bongartz, A. (2014). Sensory analysis and consumer acceptance of 140 high-quality extra virgin olive oils. *Journal of the Science of Food and Agriculture, 94,*2124–2132.50. Veneziani, G., Esposto, S., Minnocci, A., Taticchi, A., Urbania, S., Selvaggini, R., Sordini, B., Sebastiani, L., Servili, M. (2018). Compositional differences between veiled and filtered virgin olive oils during a simulated shelf life. *LWT - Food Science and Technology, 94,* 87–95.

51. Zullo, B.A., & Ciafardini, G. (2018). Changes in Physicochemical and Microbiological Parameters of Short and Long-Lived Veiled (Cloudy) Virgin Olive Oil Upon Storage in the Dark. *European Journal of Lipid Science &. Technology,120,* http://doi.org/10.1002/ejlt.201700309

52. Xenakis A., Papadimitriou, V., & Sotiroudis, T. G. (2010). Colloidal structures in natural oils. *Current Opinion in Colloid & Interface Science, 15,* 55–60.

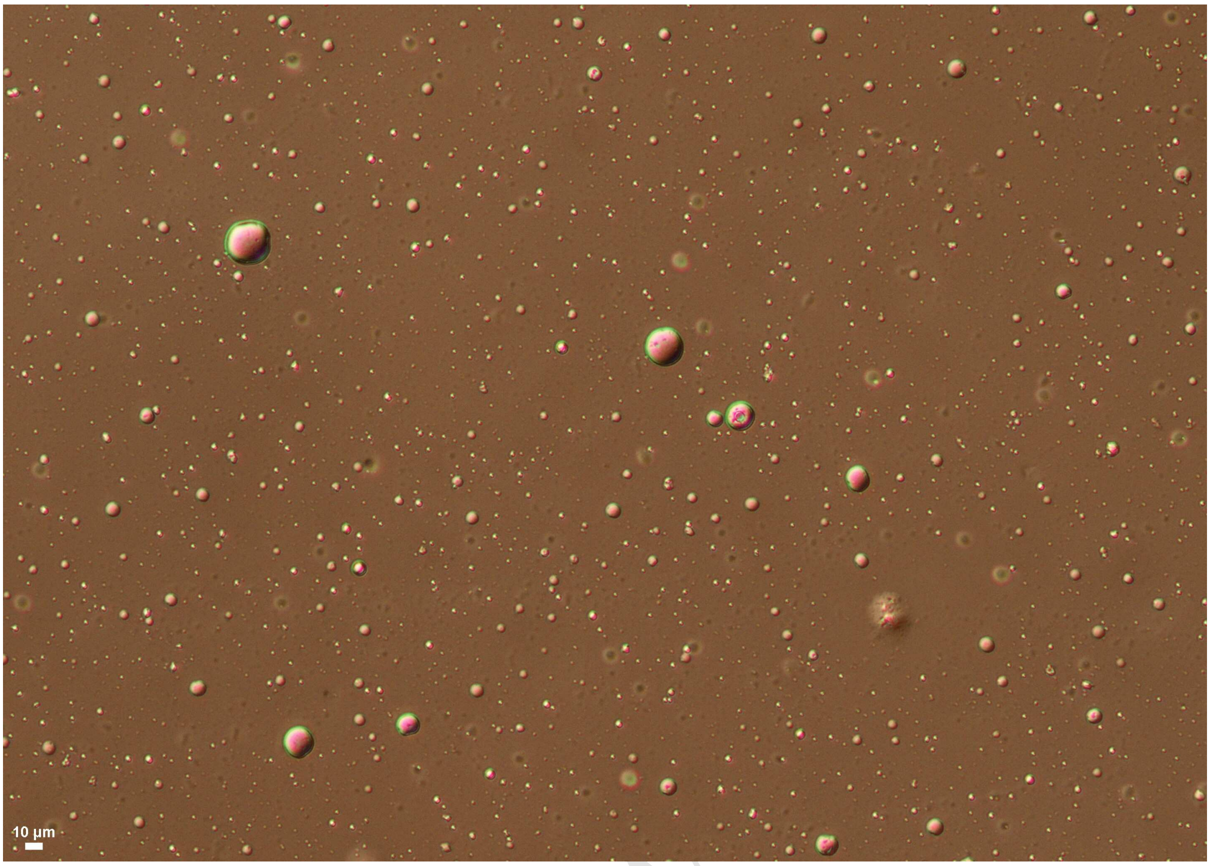
Figure captions

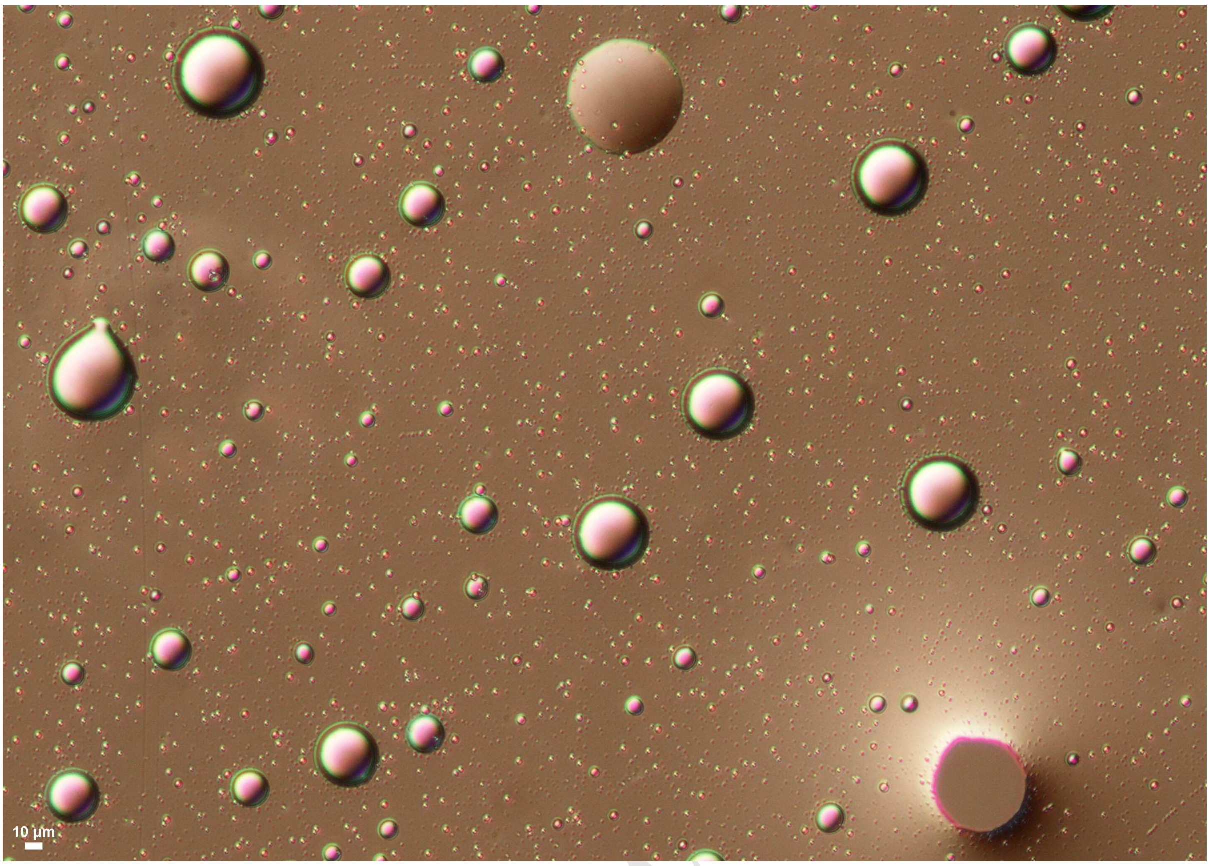
Figure 1. Virgin olive oil original examined with 10X objective.

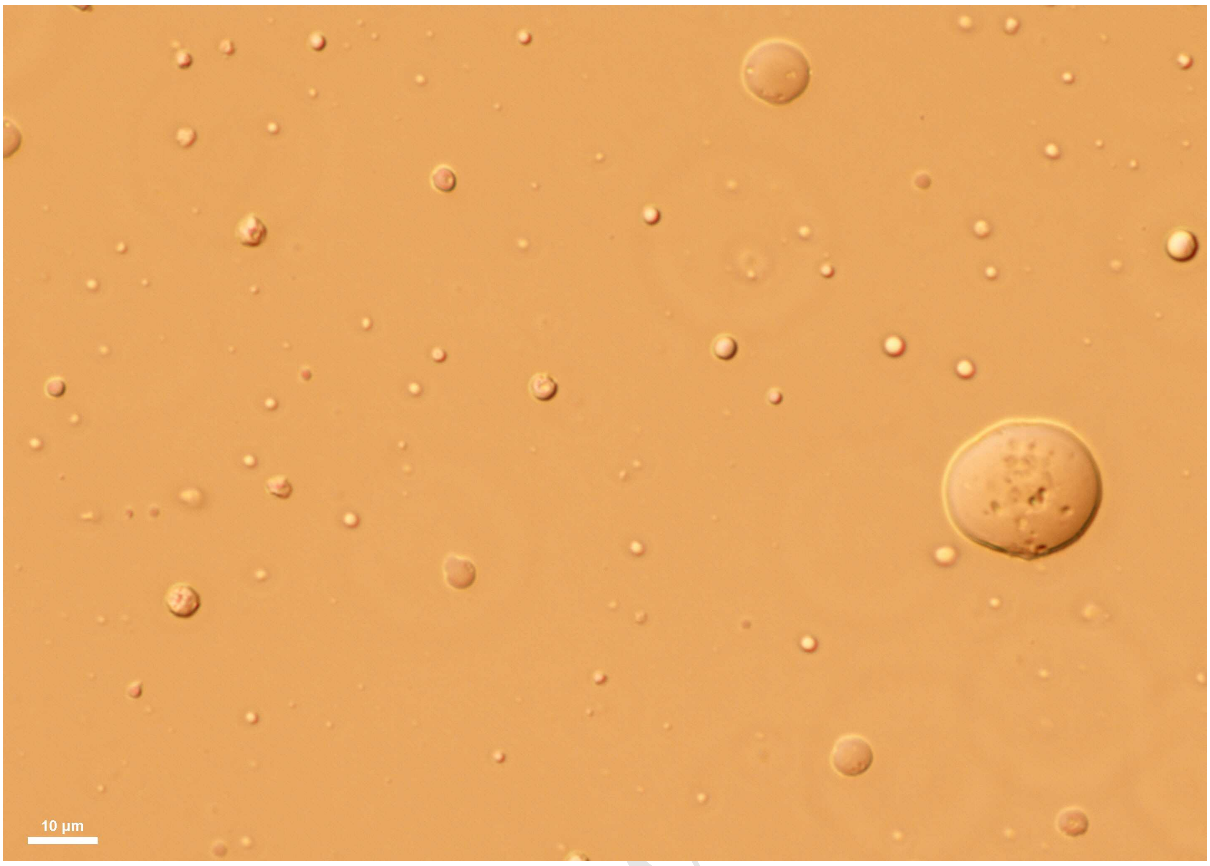
Figure 2. Virgin olive oil homogenized with water 20% v/v examined with 10X objective.

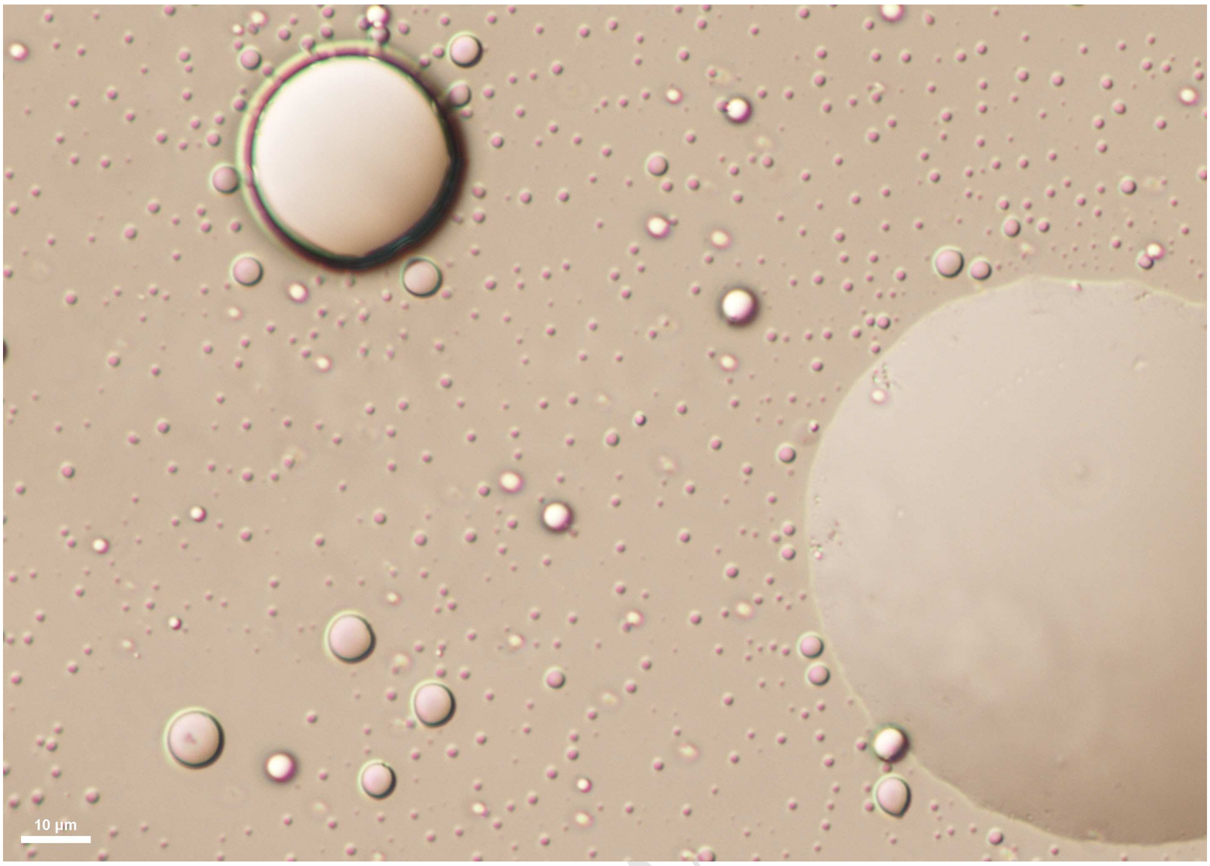
Figure 3. Virgin olive oil original examined with 40X objective.

Figure 4. Virgin olive oil homogenized with water 20% v/v examined with 40X objective.


10 µm


10 µm


10 μm


10 µm

Highlights

Fresh virgin olive oils constitute a complex colloid emulsion-sol

The new concept 'colloidal stability' is suggested

Colloidal water bags of veiled virgin olive oil are shown as characteristic

Moving particles contained in the colloidal water bags are shown