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Development of a Measure Cell to Assess by Amperometry Dissolved Oxygen in Vegetable Oils

Ahmad Ben Fakhri, Jacques Artaud, Gregory Cano, and Philippe Moulin*

Dissolved oxygen in vegetable oils is an important component of oil degradation over time. Various methods for the determination of dissolved oxygen in oils are proposed in the literature leading to very variable results. An amperometric method associated with a cell of measurement in dynamic mode makes it possible to determine the conditions of stability of the measurements of the relative dissolved oxygen concentration (RDOC, mg L⁻¹) and the electrolysis current intensity (ECI) (nA) in seed oils and virgin olive oils. The effects of oxygenation/deoxygenation time and temperature on RDOC measurements are investigated. For virgin olive oils, the RDOC in bottles closed for eight weeks varies from 0 to 0.9 mg L⁻¹. The nature of the fruitiness and the filtration of the oils have no influence on the RDOC. The monitoring of RDOC and ECI during the flow of virgin olive oil in a column open to air, simulating an industrial tank, shows differences in dissolved oxygen concentration due to a faster flow of oil in the center of the column than at the walls. This result is validated by a similar experiment with nitrogen inerting the column and by a numerical simulation.

Practical applications: Knowledge of the oxygen dissolved in vegetable oils is an important factor in limiting the autoxidation phenomenon to increase their shelf life. The development of a cell for measuring the relative dissolved oxygen concentration (RDOC) in vegetable oils and its conditions of use, provides a tool for the RDOC applicable to the different stages of the manufacturing process and during storage or packaging. The RDOC measurement is particularly useful during long storage in tanks whether inerted or not.


1. Introduction

The quality of virgin olive oil (VOO) depends on many factors that occur from lipogenesis to consumption. Some factors are very important such as the sanitary condition of olives, the time between harvesting and extraction, the process to obtain the oil, storage and packaging. Oxygen has two antagonistic roles, one positive during mechanical extraction process (lipoxygenase pathway)^[1] and the other negative during storage (oxidation).^[2] Although VOO has antioxidant compounds (phenolic compounds, tocopherols, etc.), mono (oleic acid) and polyunsaturated (linoleic and linolenic acids) fatty acids in VOO make it sensitive to oxidation according to two different processes: photooxidation and autoxidation.^[3–5] Autoxidation is the main factor in the degradation of oils. It leads to a deterioration of sensory properties with the appearance of defects such as rancidity. This defect can lead to downgrading of the oil, which can make it unfit for consumption (virgin lampante), which constitutes an economic loss for producers and distributors. During storage, the oil oxidation can be due to the oxygen present in the head space of containers or storage tanks^[6–9] and to dissolved oxygen during

the extraction process.^[10] These authors concluded that the centrifugation step at the end of the extraction process, contributed the most to the dissolved oxygen content in VOO. Oxygen quantification methods in foods and vegetable oils were reviewed by Pénicaud et al.^[11] Few data for dissolved oxygen in vegetable oils are available in literature.^[12] Moreover, values given were very variable due to the measurement methods used and many parameters which can interfere with these measurements (type of samples, storage period, type of packaging, etc.).^[10,12–18] Among the different methods used to measure dissolved oxygen in VOO, the electrochemical method (Clark's electrode) was used by Italian authors.^[10,17–20,21] The electrochemical method only gives the value of the chemical potential of oxygen in the oil and its variations, but not oxygen concentration.^[11] The electrolysis current intensity (ECI) measurement has the advantage over the relative dissolved oxygen concentration (RDOC) measurement of having a better precision on the variation of the measurement (tenth of a nano-ampere) but has the disadvantage that the link between chemical potential and dissolved oxygen concentration is not

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obvious. The operating protocols are not sufficiently described in the literature, particularly on two points: calibration and stability of the measurement over time. The aim of this study is to develop a cell for measuring chemical potential, function of the dissolved oxygen concentration in virgin olive oil, and to determine the operating conditions to obtain a stabilized measurement. This methodology was applied to the measurement of dissolved oxygen in seed oils and in virgin olive oils as well as to the oxygen concentration during the flow of an oil in a column in contact with air.

2. Experimental Section

2.1. Oil Samples

Details of the samples, used in this study, are:

- three commercial vegetal oils, purchased in April 2021: peanut (1 L), rapeseed (1 L), and sunflower (1 L);
- ten virgin and extra virgin olive oils (1.5 L) including five French Protected Designation of Origin (Corse, Nice, Nyons and Vallée des Baux de Provence) and five varietal mixt from French departments (Bouches du Rhône, Gard, Var and Vaucluse) (2020), provided by the Centre Technique de l'Olivier (CTO, France Olive, Aix-en-Provence, France);
- 5 L of VOO (varietal mixt—harvest 2019–2020 and 2020–2021) provided by the CTO,
- 4 L of VOO (cultivar Bouteillan) from 2017 to 2018 (Var, France) and
- 1 L of VOO (cultivar Bouteillan) from 2020 to 2021 (Var, France).

Sample selection could be different by incorporating a more saturated oil such as palm oil^[12] where oxygen was more soluble but we have chosen unsaturated oils which are the most sensitive to oxidation.

2.2. Experimental Design

The experimental plant included an oil tank (3 L separatory funnel or 2 L column) connected to a measuring cell (73 mL) (IMECA, Montpellier, France) (Figure 1). The column (height = 50 cm, diameter = 6.5 cm) was equipped with a tap in its lower part (orifice diameter = 0.7 cm) and could contain 1.5 L of oil. It simulated the flow of a storage tank. In its upper part, an opening allowed to adapt the measuring probe (Clark electrode) (InPro 6850i/12/120, Mettler Toledo) connected to a transmitter (M400, Mettler Toledo), fits. The cell was equipped with two openings allowing the oil feed (bottom part) and the oil outlet (lateral middle part). The oil mass flow (kg h^{-1}) was measured with a balance and a chronometer. The transmitter allows the polarization of the electrodes of the probe (600–800 mV). It indicates the electrolysis current intensity of (ECI) flowing between the two electrodes (nA) (silver anode, platinum cathode), the temperature of the measurement ($^{\circ}\text{C}$), and a relative dissolved oxygen concentration (RDOC, mg L^{-1}). The measurements were carried out in an air-conditioned laboratory (24°C). Prior to the measurements, the electrode was calibrated^[18] in air (maximum intensity -61.6 nA) and also in the work, in nitrogen (minimum intensity

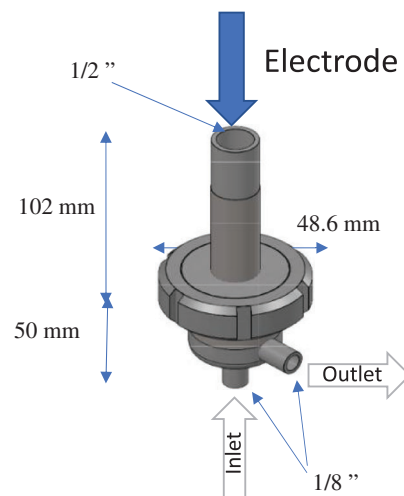


Figure 1. Cell with housing for Clark electrode by IMECA (Montpellier, France) (InPro 6850i/12/120, Mettler Toledo).

-0.2 nA). Since the calibration was done against air, the oxygen concentration value has only a relative value and did not correspond to the real value of dissolved oxygen. In this article, the ECI and RDOC will be used. The measurements were performed with 0.5–3 L of oil depending on the tank used which was loaded above the cell. The mass flow rate, ECI, temperature, and RDOC were recorded periodically (30 s). Three types of measurements were successively performed: i) at the opening of the oil bottles (OB), ii) after oxygenation by bubbling air (OX), and iii) after oil degassing by bubbling nitrogen (N_2). Oxygenation and degassing are performed in cylindrical "bubblers" (height = 20 cm, diameter = 14 cm) equipped with a glass porous medium (porosity 4) in its lower part and a lateral tube allowing an air or nitrogen supply (5 L min^{-1}).

2.3. Numerical Simulations

Numerical simulations were made with Simcenter Star-CCM+ software (Siemens Digital Industries Software, 92320, Chatillon, France) using the finite volume method where the Navier–Stokes equations are solved using a high-precision computer for a laminar flow. Polyhedral cells were used for the mesh allowing a better control over the refinement grid with an optimization of the cells.

3. Results and Discussion

The determinations of the ECI and RDOC in oils require the study of the different operating parameters of the experimental device to obtain constant and stable measurements. The measurements were performed dynamically which required: i) the determination of the time to obtain a stable amperage measurement; ii) the influence of the oil mass flow rate in the cell; and iii) the optimal oxygenation and deoxygenation times of the oils. The optimized conditions will be applied to the measurements of ECI and RDOC in various vegetable oils. These two values will be used and discussed to take into account the precision of the first and to have an idea of the order of magnitude of the relative concentration of dissolved oxygen for the second.

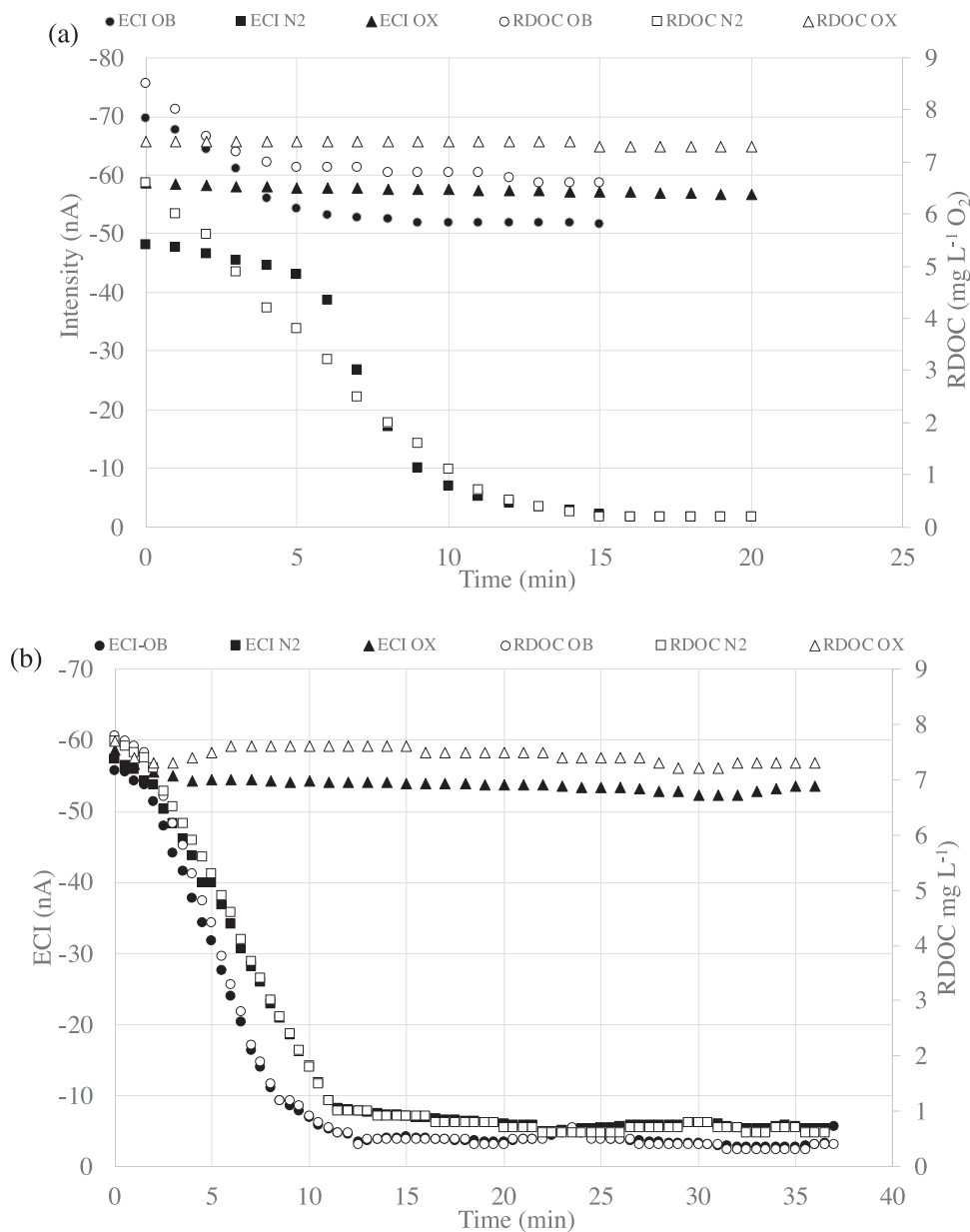


Figure 2. Variations of ECI and RDOC as a function of time for two different virgin olive oils: a) Bouteillan 2020–2021; b) Bouteillan 2017–2018, $T = 24\text{ }^{\circ}\text{C}$; OB: bottle opening, N_2 : degassing by bubbling nitrogen, and Ox: oxygenation by bubbling air.

3.1. Optimization of Experimental Conditions

3.1.1. Determination of the Stabilization Time of the ECI and RDOC Measurements

To determine the time required to obtain a stable measurement of ECI and RDOC, two virgin olive oils (Bouteillan) were used. The measurement was first performed after opening the bottle. Then, the measurement was repeated after the oil deoxygenation by bubbling nitrogen for one night (12 h), then after the oxygenation by bubbling air for one night (12 h). The aim was to determine if the stabilization time is function or not of on the oxygen

concentration. **Figure 2** shows the results for two oils from two different harvest years.

In **Figure 2a**, virgin olive oil (OB) initially contains oxygen. These values were stabilized after 15 min (ECI: -51.7 nA and RDOC: 6.6 mg L^{-1}). The intensity variation being more sensitive than the variation of dissolved oxygen, stabilization was taken into account after three consecutive constant intensity measurements ($1\text{ measurement min}^{-1}$). After deoxygenation by N_2 , the two values decreased differently but both stabilized after 15 min around -1.8 nA and 0.1 mg L^{-1} for ECI and RDOC, respectively. After oxygenation of the oil by air (OX), the stabilization appears between 15 and 20 min with ECI and RDOC values of

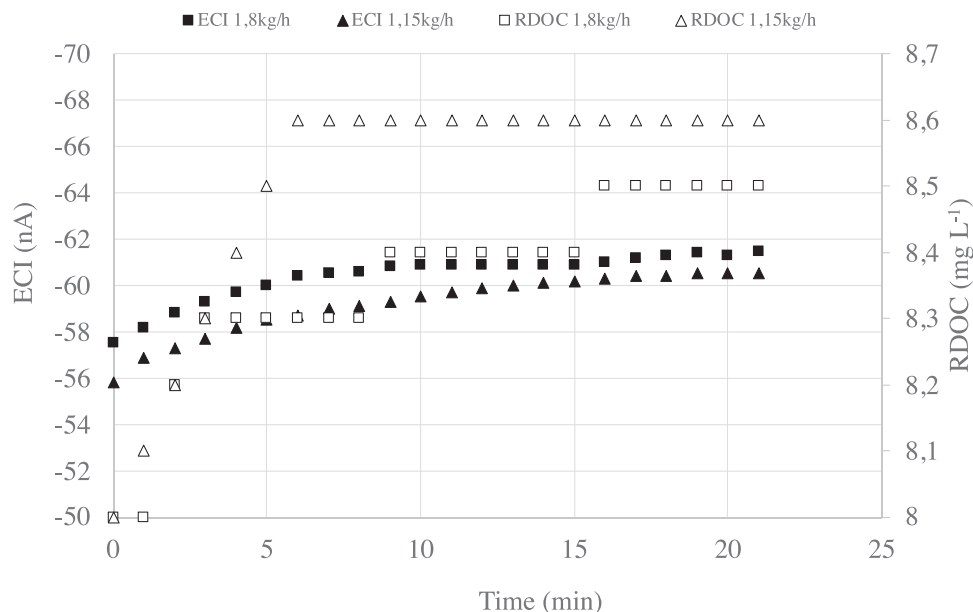


Figure 3. Variations of ECI and RDOC as a function of time for two different oil flow rates (laminar flow; rapeseed oil).

−51.7 nA and 7.4 mg L^{-1} , respectively. In Figure 2b, for an older oil (Bouteillan 2017–2018), identical stabilization times were obtained. However, the figure shows that at the opening of the bottle (OB) the RDOC is very low probably caused by the consumption of oxygen by polyunsaturated fatty acids of the triacylglycerols contained in the oil.^[22] The stabilization time will be fixed between 15 and 20 min for this apparatus and the protocol used.

3.1.2. Effect of the Flow Rate

The influence of the flow rate in the cell measurement was studied for two flow rates of 1.8 and 1.15 kg h^{-1} (Figure 3). These flow rates correspond to a laminar regime in the measuring cell and to a sufficient shear stress in the vicinity of the electrode to renew the oil and allow obtain a limited oil consumption for the measurement (around 500 g). To measure the effect of the flow rate, it was important that ECI and RDOC were constant and same, so the oil is oxygenated by air for 4 h before each experiment (i.e., oil has been saturated). The ECI measurement appears more sensitive because it constantly evolves over time, whereas the RDOC measurement evolves in steps and in a measuring range narrower than that of the ECI. The choice to consider the ECI as a stabilization parameter was justified. For the two flows studied, stabilization appeared after 20 min and the stabilized values were identical. In the following measurements, a flow rate of 1.8 kg h^{-1} with an oil consumption of 500 g per measurement has been chosen.

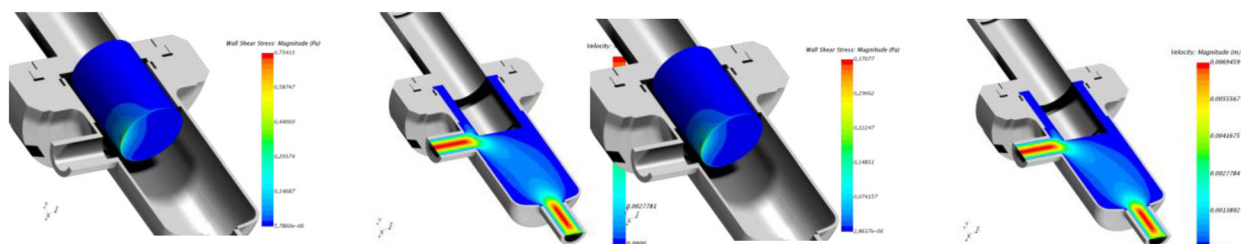
The shear stress in the measuring cell was determined by computational fluid dynamics for these two flows. A polyhedral mesher has been used to generate 450 000 cells. It is relatively easy and efficient to build, requiring no more surface preparation than the equivalent tetrahedral mesh. Regarding the physical model, a stationary laminar flow has been chosen consider-

ing the effect of gravity, density, and dynamic viscosity of olive oil. For boundary conditions, a mass flow rate is imposed at the inlet, and a pressure outlet is used as an outflow condition which can be considered as the static pressure of the environment into which the fluid enters. Finally, a no-slip condition is used on the walls as we want to evaluate shear stress on the oxygen sensor.

Velocity fields and values of the shear stress on the surface of the cell for these two flows are given in Figure 4, respectively, 1.396×10^{-2} and $6.96 \times 10^{-3} \text{ Pa}$. The numerical simulation shows a laminar regime with current lines (not shown on Figure 4) which validated the absence of dead volume and more especially at the electrode wall. In this case, i) the electrode is always in contact with moving oil and does not measure values for stagnant oil and ii) in the cell, the oil is always moving so the flow is not disturbed and there is no under, or oversaturated oxygen area compared to the circulating oil.

3.1.3. Effect of Oxygenation/Deoxygenation Time

The development of an analytical technique for the measurement of dissolved oxygen in vegetable oils requires the ability to measure concentrations at saturation but also close to zero. For this purpose, bubbling with air or nitrogen was, respectively, carried out in 2 L reactors, containing 1 L of oil equipped with a porous media (porosity 4) in its lower part allowing a supply of air or nitrogen. Nitrogen bubbling times of 1 h 30 min, 4 h, and 17 h were tested (Figure 5). In this time range, the degassing time had no importance on the final RDOC which stabilized after 20 min—around 0.8 mg L^{-1} . However, Figure 5 shows that the degassing time of 1 h 30 min leads to a slower drop in the dissolved oxygen concentration and is different from the two others. For this study, a duration of 4 h will be used for the oxygenation or deoxygenation phases.



Shear stress = $1,396 \cdot 10^{-2}$ Pa

Shear stress = $6.96 \cdot 10^{-3}$ Pa

Figure 4. Determination of the shear stress in cell by numerical simulation for two different flow rates (left, 1.8 kg h^{-1} ; right, 1.15 kg h^{-1}).

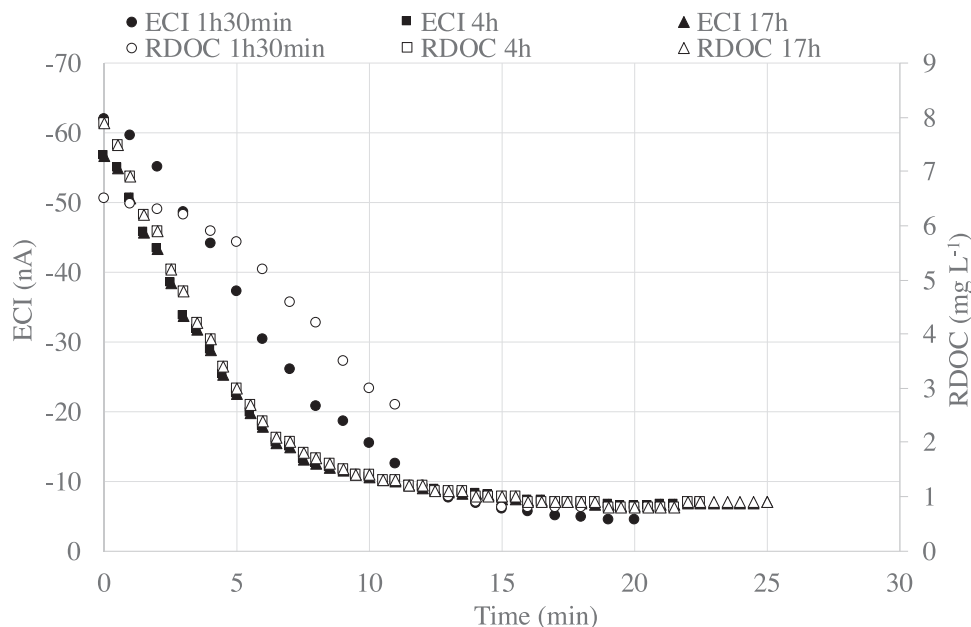


Figure 5. Variations of ECI and RDOC as a function of time for three different degassing times (rapeseed oil).

3.2. Temperature Effect

The oxygen solubility in oil was studied for three experiments at two temperatures (14 and 28 °C) for an air (oxygen) saturated oil obtained by bubbling during 4 h in a thermostatic bath. The first experiment at 14 °C (**Figure 6a**) shows that starting from an ambient temperature of 22 °C, the temperature decreases. After 12 min, stabilization of RDOC was obtained around 9.2 mg L^{-1} . Without temperature control, the temperature of oil increased to a final value around 17 °C and a RDOC value of 8.4 mg L^{-1} . This figure highlights the accuracy of the measurement since the temperature decreases and then increases slightly, which results in an inverse variation for the RDOC. Also, for a temperature of 16.5 °C obtained in increasing and decreasing periods of temperature, the RDOC was similar in both cases around 8.8 mg L^{-1} . **Figure 6b** shows the results at 28 °C for a stabilized temperature of 27 °C. This experiment was repeated twice to test the repeatability and to consider the fact that the pilot and more especially the cell were not thermostated. The results were repeatable and the RDOC for these two experiments is similar: $6.5 \pm 0.1 \text{ mg L}^{-1}$.

These results show the RDOC at air saturation in oil as a function of temperature vary in a similar way to the oxygen concen-

tration in water: when the temperature increases, the RDOC decreases. For temperatures of 14.8, 22, and 28 °C, RDOCs of 9.2, 7.3 (**Figure 2b**), and 6.4 mg L^{-1} were obtained, respectively.

3.3. Effect of Various Seed Oils and Virgin Olive Oils

3.3.1. Vegetable Seed Oils

Experiments have been realized on three commercial vegetable oils (peanut, rapeseed, and sunflower). When the bottles were opened, the dissolved oxygen concentration was very different with RDOC of 3.7, 6, and 0.7 mg L^{-1} , respectively, for peanut, rapeseed, and sunflower oils (**Figure 7**). After oxygenation by air and degassing by N_2 , an identical trend appears for the three oils, namely, a stabilization RDOC around 8–9 and 0.7 mg L^{-1} , respectively.

3.3.2. Virgin Olive Oils

Table 1 gives the nature of the fruitiness (green, ripe, and black) of virgin olive oils, the filtration or absence of filtration of the oils

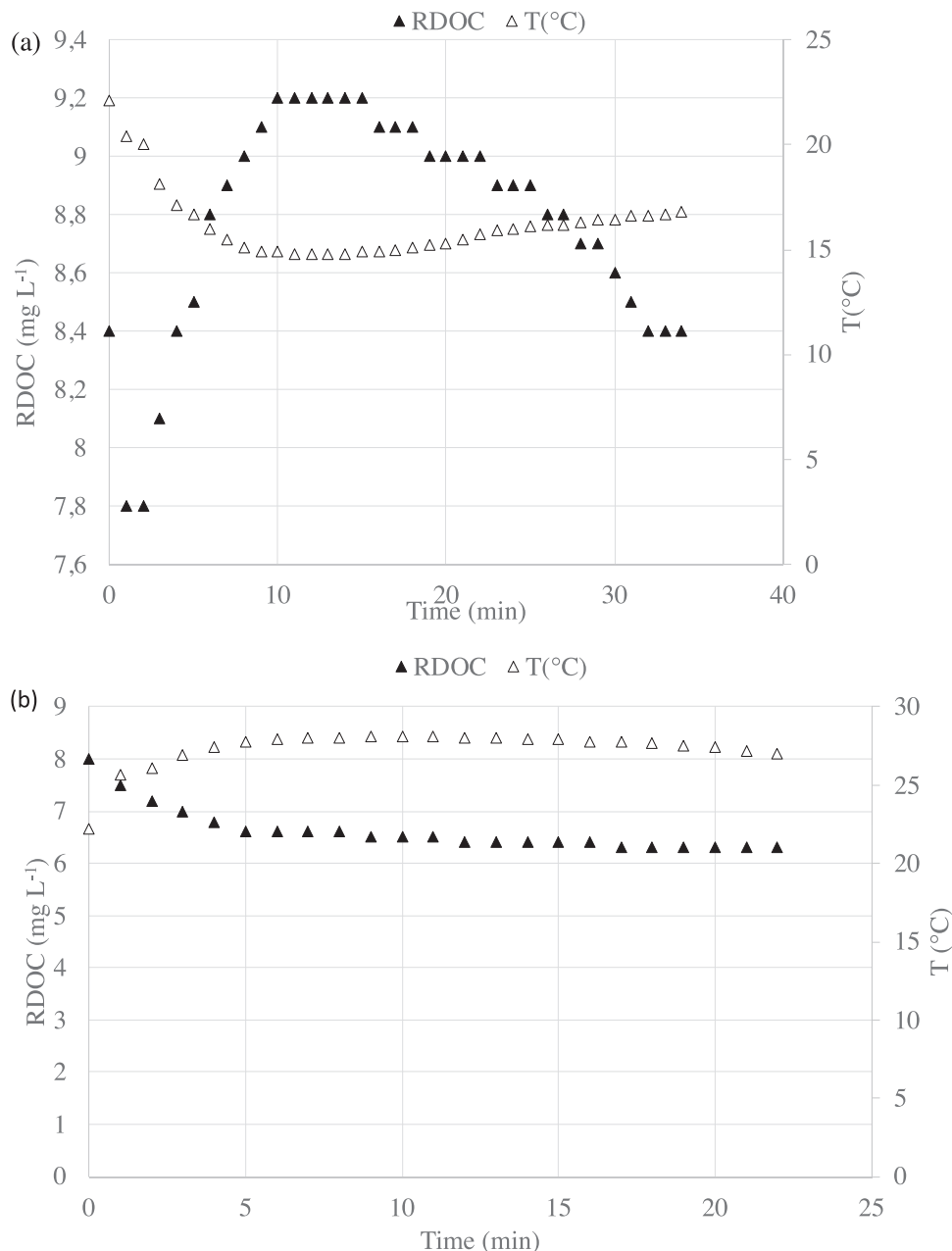


Figure 6. Variations of RDOC and temperature as a function of time: a) low temperature: 14 °C; b) high temperature: 28 °C (Bouteillan, 2017).

as well as the RDOC when the bottles were opened and after deoxygenation by bubbling nitrogen or oxygenation by bubbling air. The green and ripe fruitiness are defined by International Olive Council^[23] and black fruity was a French specificity. For black fruitiness olive oils, the olives are subjected to controlled anaerobic fermentation in closed bin (320 kg) for a few days before the oil extraction. When the bottles were opened (OB) (Table 1), whatever the nature of the fruitiness and the filtration or absence of filtration of the oils, the RDOC was relatively low and did not exceed 0.8 mg L⁻¹. After a 4 h degassing (N₂), all the values remained low. These values can be slightly higher than at the bottle opening. This can be explained by the order of the experiments

after the bottle opening (OX-N₂ or N₂-OX). This experimentation order was random and depends on the availability of bubblers. Thus, an oxygenated oil by air after opening the bottle and then undergoing a 4 h degassing with nitrogen does not allow to obtain a RDOC as low as the natural consumption process during a long term. On the other hand, after oxygenation by air (OX), the RDOC remained approximately the same for all the oils analyzed, around 7.3–7.4 mg L⁻¹ except for the oil of Corsican origin which reached 8 mg L⁻¹. In conclusion, these results show that the nature of the fruitiness with or without filtration of the VOO did not appear to have any effect on the RDOC at saturation or in the absence of oxygen.

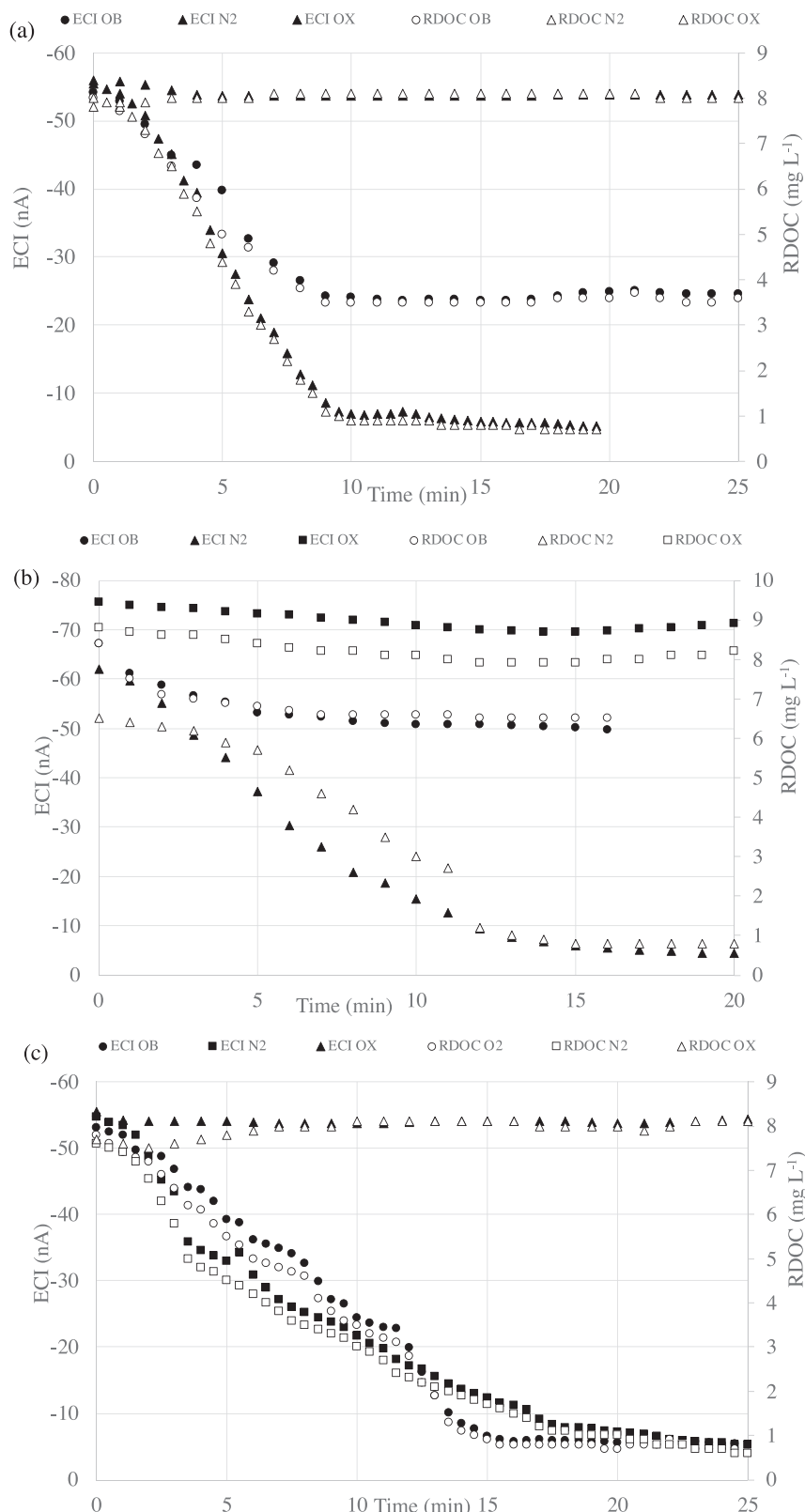


Figure 7. Variations of ECI and RDOC as a function of time for three different vegetable oils: a) peanut; b) rapeseed; c) sunflower. OB: bottle opening, N₂: degassing by bubbling nitrogen, Ox: oxygenation by bubbling air.

Table 1. RDOC for different virgin olive oil origins and different producers.

Origins	Fruity	Filtered	RDOC		
			OB	N ₂	OX
Corse PDO (20)	Ripe	Yes	0.0	0.3	8.0
Gard (30)	Ripe	No	0.7	0.7	7.4
Nice PDO (06)	Ripe	No	0.6	0.4	7.5
Nyons PDO (26)	Ripe	Yes	0.5	0.1	7.4
Bouches-du-Rhône (13)	Black	Yes	0.1	1.0	7.2
Bouches-du-Rhône (13)	Green	Yes	0.0	0.6	7.4
Vallée-des-Baux de Provence PDO (13)	Black	Yes	0.8	0.6	7.4
Vallée-des-Baux de Provence PDO (13)	Green	Yes	0.7	0.8	7.4
Var (83)	Black	No	0.5	0.6	7.3
Vaucluse (84)	Green	Yes	0.4	0.2	7.7

Notes: RDOC: relative dissolved oxygen concentration, $T = 24\text{ }^{\circ}\text{C}$ OB: bottle opening. N₂: degassing by bubbling nitrogen. OX: oxygenation by bubbling air (n): identification number of French departments

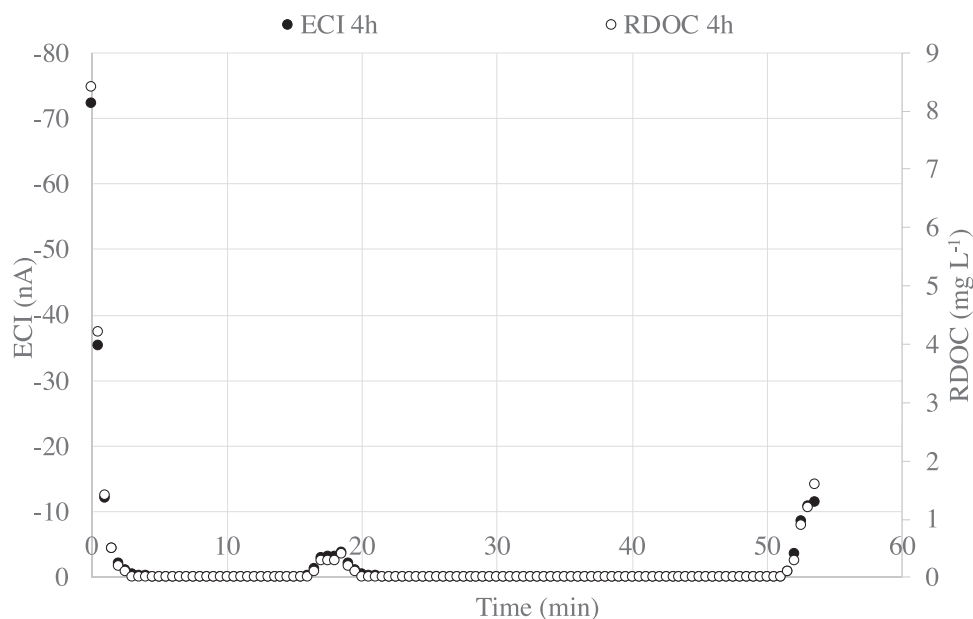


Figure 8. Variations of ECI and RDOC: dynamic measurement (CTO, varietal mix 2020–2021).

3.3.3. Dynamic Measurement: Simulation of the Flow from an Industrial Tank

This experiment was carried out using a column that can contain 1.5 L of oil and simulates the flow of an industrial tank. The VOO (CTO, varietal mixture 2020–2021) was previously degassed for 4 h before being gently poured into the column. **Figure 8** shows that the RDOC and the ECI rapidly decrease to zero. A slight increase in RDOC and ECI was observed after 16–17 min before returning to zero. This time corresponds to approximately one-third of the total flow time. When the flow was finished, the entry of air into the measuring cell causes an increase of RDOC and ECI. The increase in the RDOC and the ECI at one-third of the flow can be explained by the presence of small quantities of oxygen present in the oil in contact with the air at the upper in-

terface which reaches rapidly the probe due to a faster flow in the center of the column than at the walls. To verify this assumption, two different approaches were carried out: a measurement of the RDOC and the ECI in an oil degassed with nitrogen and placed in a totally inerted column during the flow and a numerical study of the flow. With an inerted column (i.e., VOO previously degassed for 4 h and nitrogen continuous injected at the top of tank), the RDOC and the ECI no longer show an increase at one-third of the emptying time of the column and no longer an increase at the end of the flow, which validates the hypothesis of the presence of oxygen due to air solubility at the air-oil interface. The numerical study of the flow showed that the oil at the interface, which is initially at the top of the column ($h_{\text{total}} = 50\text{ cm}$) in contact with the ambient air, crossed the drain orifice of the column before the oil located near the walls, thus explaining

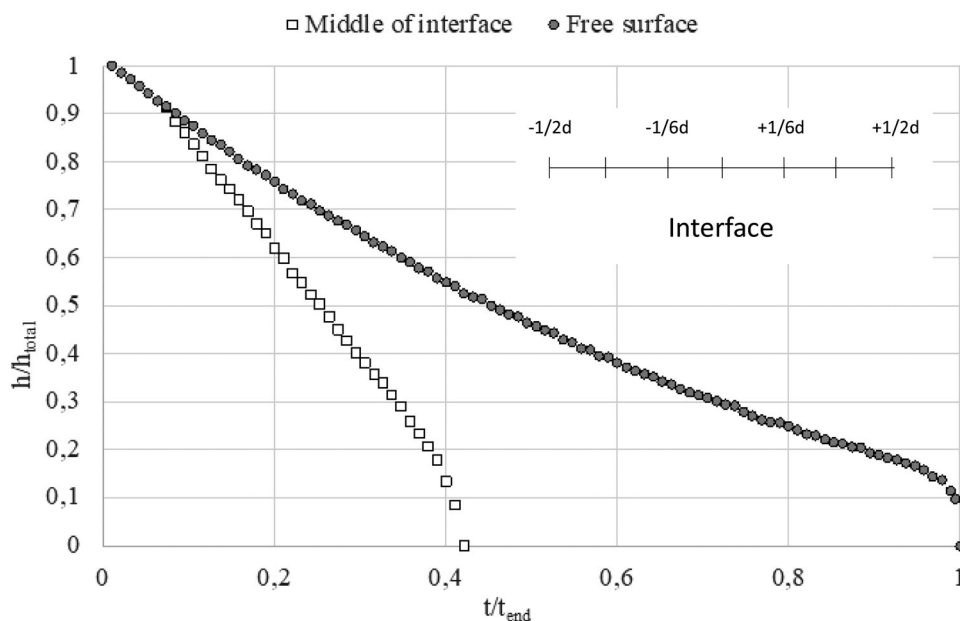


Figure 9. Variation of the ratio of the dimensionless height h/h_{total} of the fluid particles initially ($t/t_{\text{end}} = 0$) located at the center of the interface (third of the diameter – middle of the interface) and on the rest of the interface as a function of the ratio (t/t_{end}), (Middle of the interface: $-1/6d$ to $+1/6d$). Rest of the interface: $(-1/2d$ to $-1/6d$ and $+1/6d$ to $+1/2d$) (t_0 is the initial time when the oil starts to flow, t is time during flow, t_{end} is the final time when the wall oil has run out, h is the height of the oil in the column at any time, and h_{total} is the height of the oil at the initial time).

the slight increase in the parameters measured. Regarding the gravitational flow in the column, the multiphase volume of fluid was used as a suitable physical model to simulate the flows on numerical grids capable of resolving the interface between the phases of the mixture. In addition, a Lagrangian phase was introduced with massless particles to quantify the residence time of the fluid in the continuous phase. Cells ($n = 750\,000$) were generated for the mesh. Computational fluid mechanics showed that under these operating conditions, the first oily element of the interface arrives for a time equivalent to 42% of the total flow time. This slight difference between simulation and experiment is explained by the fact that the experimental measurements are made in the cell after the column while the simulation gives the values at the outlet of the column hence the time lag. **Figure 9** shows, as a function of ratio (t/t_{end}), the variation of the ratio of the dimensionless height h/h_{total} of the fluid particles initially ($t/t_{\text{end}} = 0$) located at the center of the interface (third of the diameter – middle of the interface) and on the rest of the interface. The sedimentation rate of particles in the middle of the interface is almost 2.5 times faster than other particles. This value of 2.5 agrees with the draining time (16–17 min) of the oil at the center of the interface and with the total draining time of the column (51 min). This result shows the importance of inerting the top of the tanks to reduce the RDOC of the oils and thus improve their conservation.

4. Conclusion

A cell for measuring dissolved oxygen in vegetable oils has been developed. The amperometric measurements in dynamic mode do not allow to obtain the dissolved oxygen concentration but a relative dissolved oxygen concentration (RDOC) because the cali-

bration of the amperometric probe was carried out in air (saturation) and in nitrogen (zero). Different operating parameters were specified in order to obtain some stable measurements: duration 20 min and flow rate 1.8 kg h^{-1} . The amperometric measurement brings a higher sensitivity and reliability of the measurement since the calibration is carried out on this parameter and the accurate (10^{-2}) is more important. RDOC were determined on seed oils and virgin olive oils. Filtration and the nature of the fruitiness of virgin olive oils do not influence the measurement. The influence of temperature on the RDOC is similar to that described for water: the RDOC decreases with temperature. The measurement of RDOC during the continuous flow of a nitrogen-deoxygenated virgin olive oil, contained in a column open to air, shows differences in RDOC due to a different velocity between the center of the tube and the walls. This analytical technique of measurement in dynamic mode can be used to follow the RDOC of an oil during the different stages of oil extraction as well as during storage or emptying of a tank.

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Conflict of Interest

The authors declare no conflict of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Keywords

dissolved oxygen, electrochemical method, measure cells, seed oils, virgin olive oils

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