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Study of the Organoleptic, Physicochemical, and Antioxidant Properties of *Citrus limon* essential oils

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Abstract

This study focuses on the extraction and analysis of the organoleptic parameters, antioxidant activity, and physicochemical properties of the essential oil (EO) from the leaves of *Citrus limon* from two Algerian regions: Sidi Harb and El Hadaiek. The organoleptic analysis characterized the sensory profiles of *Citrus limon* essential oils, highlighting variations in color. The essential oil yield was also calculated for each region, revealing notable differences influenced by the specific environmental and agronomic conditions of each site. The results of the physicochemical analysis show variations between the samples from the different regions. The pH, refractive index, and Brix index were measured and compared to determine the quality of the essential oils extracted from *Citrus limon* leaves. Additionally, the antioxidant activity of *Citrus limon* essential oils was calculated, showing significant differences between the two regions.

This study provides valuable information on the physicochemical, organoleptic, and antioxidant properties of *Citrus limon* leaf essential oils, highlighting the importance of the region of origin on the quality and yield of the essential oil. This data can be used to optimize extraction methods and improve the quality of finished products intended for various industrial and food applications.

Keywords: Organoleptic parameters, antioxidant activity, physicochemical parameters, *Citrus limon*, Sidi Harb, El Hadaiek.

1. Introduction

Algeria has approximately 3,139 identified plant species, with notable diversity. About 35% of these species are of Mediterranean origin, 12% Euro-Mediterranean, and 11% North African. Additionally, 700 of these species are endemic to Algeria, highlighting the richness and uniqueness of the country's flora (IUCN, 2018). More than a quarter of the plants in Algeria are

traditionally used for medicinal purposes. Among these plants, *Citrus limon*, commonly known as the lemon tree, holds significant importance due to its numerous therapeutic benefits.

This study focuses on *Citrus limon*. This plant is not only valued for its fruits but also for its leaves and essential oil, which are widely used in various medicinal and aromatic practices. The lemon tree is particularly appreciated for its biological properties (Kačániová et al., 2024). Antioxidant activity is crucial as it helps to neutralize free radicals in the body, thereby reducing the risk of chronic and inflammatory diseases. The essential oil of *Citrus limon* has been shown to possess significant antioxidant activity, which can contribute to its therapeutic benefits. By mitigating oxidative stress, these antioxidants can protect cells and tissues from damage, promote overall health, and potentially prevent various health conditions.

This research aims to evaluate the quality, safety, and antioxidant activity of lemon essential oil to confirm its potential application in the pharmaceutical and parapharmaceutical industries.

2. Materials and methods

2.1. Regions of study

The *Citrus limon* used in this study originates from two regions in Northeastern Algeria:

Sidi Harb: Sidi Harb is a region located in the commune of Annaba, known for its Mediterranean climate, which is favorable for citrus cultivation. This area benefits from fertile soils and adequate rainfall, making it an ideal environment for growing lemon trees.

El Hadaiek: Located in the commune of Skikda, El Hadaiek is distinguished by its humid subtropical climate, influenced by its proximity to the sea. The Skikda region, in general, is recognized for its varied agricultural production, ranging from cereals to fruits, with a particular emphasis on citrus due to favorable agro-climatic conditions.

2.2 Plant material

Citrus limon, a majestic tree that can reach five meters in height, offers the world a precious fruit; the lemon. Belonging to the Rutaceae family (Gollouin and Tonelli, 2013), this citrus fruit is adorned with a thick rind, evolving from tender green to bright yellow under the influence of cold. As an emblem of the Mediterranean sun, it symbolizes health and vitality, standing out as one of the most cherished fruits around the globe (Gonzalez-Molina et al., 2010).

Prior to extraction, the plant, cultivated in Sidi Harb and El Hadaiek regions, was rinsed with distilled water and air-dried in the shade at room temperature for 10 days to preserve the integrity of its molecules. The dried leaves and stems were then ground into powder using an electric grinder. The resulting powder was sieved and stored away from light until use.

For the hydrodistillation extraction method employed in this study, 140 g of dried plant material was placed in a 1-liter ballon flask of a Clevenger apparatus, with an appropriate amount of distilled water added (2/3 of the flask volume). Extraction was carried out for two hours from the onset of boiling. Finally, the obtained essential oil was stored at 4°C. The quantity of oil obtained was measured to calculate the extraction yield.

2.3. Organoleptic and Physicochemical properties of *Citrus limon*

Essential oils have common sensory properties, including characteristics that can be perceived by the sensory organs: color, odor, flavor or taste, and appearance or aspect. For this purpose, we provided several samples of *Citrus limon* essential oil from each region to 50 persons to determine their organoleptic properties immediately after extraction and recovery. The essential oil of *Citrus limon* is subjected to a series of physicochemical analyses, including pH, refractive index and Brix index.

2.4. Antioxidant activity of *Citrus limon*

The antioxidant activity of essential oils extracted from *Citrus limon* leaves, sourced from two regions, Sidi Harb and El Hadaiek, was evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay. The DPPH method was employed to assess the ability of the essential oils to scavenge free radicals, with vitamin C used as a standard for comparison. Essential oils were tested for their antioxidant properties by measuring their capacity to neutralize the DPPH radical. The results were compared to the antioxidant activity of vitamin C, which served as a reference standard, to determine the efficacy of the essential oils in reducing oxidative stress. The antiradical activity is calculated as a percentage of inhibition using the following formula (Amira et al., 2024):

$I\% = (A_0\text{-}A_s) / A_0 \times 100$

Where A_0: Absorbance of DPPH in solution (negative control or blank) and A_s: Absorbance of Samples

The IC50 (Inhibitory Concentration 50%) or EC50 (Efficient Concentration 50%) refers to the concentration of antioxidants needed to inhibit or neutralize 50% of free radicals. This value is determined graphically through linear regression of the inhibition percentages plotted against varying concentrations of antioxidants. A lower IC50 value indicates a higher antioxidant capacity of the compound (Reguez et al., 2023).

3. Results and discussion

The organoleptic parameters of our essential oils (color, odor, flavor, and appearance) are summarized in Table 1. These results are consistent with those reported in the literature (Bey Ould Si Said, 2014; Bessedik and Khenfer, 2015; Reghaissia, 2020; Gueffal and Zengui, 2021).

Table 1: Organoleptic properties of *Citrus limon* essential oil from the two study regions.

Regions	Color	Odor	Savor	Aspect
Sidi Harb	Transparent	typical of citral	bitter taste	Liquid
El Hadaiek	pale yellow	typical of citral	bitter taste	Liquid

The yield is calculated based on the weight of the extracted essential oil (EO) relative to the weight of the plant material (PM) used in the hydrodistillation. The evaluation of the essential oil extraction yield from lemon in three different regions shows interesting results. In Sidi Harb the yield is 0.21%, while in El Hadaiek, it is higher, reaching 0.3% (table 1). In contrast to our results, the extraction yield reported by Didi and Yakoubi (2021) is 2.5%. According to Bey Ould Si Said (2014), the yield of lemon essential oil is lower (0.1%) compared to our results. The obtained essential oils are 100% pure, 100% natural, and the quantity is always low relative to the plant material used. The differences in yield in the cited studies could be explained by variations in the extraction technique used (Clevenger), local production conditions, or even the moisture content of the plant organs.

The pH analysis of the lemon essential oil from the three distinct regions does not reveal notable variations. In Sidi Harb and El Hadaiek, the measured pH is 6, indicating slightly lower acidity (table 2). Our results are consistent with those of Meflah (2015), Gueffal and Zengui (2021), and Kerfi-Gueteb and Benyahia (2020). The differences in pH among the three studied regions may be attributed to several factors, such as climatic conditions, soil composition, and agricultural practices specific to each region.

The study of the refractive index of lemon essential oil from three different regions reveals significant variations. In Sidi Harb, the refractive index is 1.4789, whereas in El Hadaiek, it is 1.4532. The refractive index indicates the essential oil's ability to reflect light. This ratio is generally high and depends on the chemical composition, which increases with the length of acid chains, their degree of unsaturation, and temperature. It primarily varies with the content of monoterpenes and oxygenated derivatives. A higher monoterpene content results in a higher refractive index. In the studies by Kerfi-Gueteb and Benyahia (2020) and Gueffal and Zengui (2021), the refractive index was higher than our results, specifically 1.4750, 1.4770, and 1.4780, respectively.

For the Brix index, the recorded values are around 75.4 and 74.1 for Sidi Harb and El Hadaiek, respectively. Although the Brix index is traditionally associated with sugar content in aqueous solutions, in the context of essential oils, it can provide valuable information about the concentration, purity, and quality of the essential oil. The interest of sugar in essential oils is generally limited, except in specific cases where the presence of sugars might influence the properties of the oil for particular applications.

Table 2: Physicochemical properties of *Citrus limon* essential oil from the two study regions

Regions	pH	Refractive index	Brix index
Sidi Harb	6	1.4789	75.4
El Hadaiek	6	1.4757	74.1
AFNOR Standard	Acidic	1.4600-1.4760	

Table 3: Equation of regression and IC_{50} (µg/mL) of *Citrus limon* essential oil (EO) and Ascorbic acid

Samples	Equation of regression	R^2	IC_{50} (µg/mL)
Sidi Harb EO	Y= 0.1867x + 0.0252	0.98	267.94
El Hadaiek EO	Y= 0.2475x + 0.0109	0.99	201.97
Ascorbic acid	Y= 0.3066x + 0.0488	0.96	161.48

The antioxidant activity of *Citrus limon* essential oil was evaluated using the DPPH assay, which measures the ability of the essential oil to neutralize free radicals. The results indicate variations in antioxidant capacity among samples from different regions. The essential oil from Sidi Harb exhibited an IC_{50} value of 267.94 µg/mL (table 3). This value reflects a moderate antioxidant activity, suggesting that the oil has a decent ability to scavenge free radicals but is not the most potent among the samples tested. The essential oil from El Hadaiek demonstrated

a higher antioxidant activity with an IC_{50} value of 201.97 µg/mL. This indicates a stronger capability to neutralize DPPH radicals compared to the oil from Sidi Harb.

The IC_{50} value for Vitamin C, used as a standard reference antioxidant, was found to be 161.48 µg/mL. This suggests that while *Citrus l*imon essential oils have notable antioxidant properties, they are generally less effective than Vitamin C in scavenging free radicals. The differences in antioxidant activity among the essential oils from various regions may be influenced by several factors, including the concentration of specific antioxidants and the overall chemical profile of the essential oil can impact its effectiveness. For instance, variations in the levels of compounds such as limonene or linalool might account for differences in radical-scavenging ability. The climatic conditions, soil composition, and agricultural practices in each region may affect the essential oil's chemical composition and, consequently, its antioxidant activity.

4. Conclusion

This study investigated the extraction of *Citrus limon* essential oil from two regions between Annaba and Skikda, focusing on its organoleptic and physicochemical parameters. The analyses revealed variations in organoleptic properties, such as color, as well as in physicochemical characteristics, including the refractive index and Brix index. These differences highlight the impact of geographical and climatic conditions on the quality of the essential oil. Additionally, the antioxidant activity of the essential oil was assessed, showing differences in its ability to neutralize free radicals between the two regions. The results provide valuable insights for optimizing extraction methods and enhancing the quality of derived products.

5. References

Amira, K. Djeghader, N. E. H., Gacem, H., Delhoum, H., Meziri, F. Chaouche, R., Boughendjioua, H. and Boudjelida, H. (2024). Chapter 5: Antioxidant activity of *Thapsia garganica,* Book of Agricultural sciences unveiled: exploring the dynamics of farming and sustainability. *by* Atena Editora. https://doi.org/10.22533/at.ed.9662430045

Bessedik, M. and Khenfer, B. (2015). Study of the Antifungal Activity of Essential Oils of *Eucalyptus globulus* and *Thymus algeriensis* Against Some Phytopathogenic Fungi of Date Palm Leaves (*Phoenix dactylifera* L). Magister's Thesis, Kasdi Merbah University, Ouargla (Algeria).

Bey ould si said, Z. (2014). ABiological Activities of Essential Oils from the Leaves and Fruit of a Medicinal Plant *Eucalyptus globulus*. Magister's Thesis, University of Bejaia (Algeria).

Didi, A. O. and Yakoubi, S. I. (2021). Extraction, Analysis, and Encapsulation of Essential Oil from Lemon (*Citrus limon*) and Orange (*Citrus sinensis*) Waste for Valorization. 81p.

Gollouin, F. and Tonelli, N. (2013). From Fruits and Edible Seeds of the World. Edition Brigitte Peyrot Poos, Paris, La voisier SAS.PP. 186-195pp.

Gonzalez-molina, E., Dominguez-perles, R., Moreno, D. A. & Garcia, V. (2010). Natural bioactive compounds of citrus limonfor food and health. *Journal of Pharmaceutical and Biomedical Analysis*, 51: 327-345pp.

Gueffal, S. and Zengui, M. (2021). Valorization of Essential Oils from Lemon Leaves. Magister's Thesis, Specialization: Petrochemical Engineering, Kasdi Merbah University, Ouargla. 59p.

IUCN: International Union for Conservation of Nature. (2024). Expert Evaluation Group: Maghreb. Available on: IUCN Green List Home - IUCN Green List, (Last updated in 2018).

Kačániová, M., Čmiková, N., Vukovic, N. L., Verešová, A., Bianchi, A., Garzoli, S., Ben Saad, R., Ben Hsouna A., Ban, Z. and Vukic, M. D. (2024). *Citrus limon* Essential Oil: Chemical Composition and Selected Biological Properties Focusing on the Antimicrobial (In Vitro, In Situ), Antibiofilm, Insecticidal Activity and Preservative Effect against *Salmonella enterica* Inoculated in Carrot. *Plants (Basel).* 13(4) : 524p.

Kerfi-Gueteb, I. and Benyahya, R. (2020). Physicochemical Characteristics of Essential Oils from Lemon (*Citrus limon*) Peel and Evaluation of Their Antioxidant and Anti-Inflammatory Activities. Magister's Thesis in Biotechnology and Plant Valorization, Department of Biotechnology, Faculty of Natural and Life Sciences, Saad Dahleb University of Blida, 55p.

Reghaissia, I. (2020). Extraction and Characterization of Essential Oil from Eucalyptus globulus: Application as an Insecticide. Magister's Thesis, Specialization: Chemical Engineering, University of 08 May 1945, Guelma. 93p.

Rguez, S., Bettaieb Rebey, I., Bourgou, S., Hammami, M. and Hamrouni Sellami, I. (2023). *Pistacia lentiscus* extracts as a valuable source of antioxidant compounds. *Journal of new sciences, Agriculture and Biotechnology*, 91 (3), 5160-5167pp.