

Soil parameters and bioclimatic characteristics affecting essential oil composition of leaves of *Pistacia lentiscus* L. from València (Spain)

Parámetros del suelo y características bioclimáticas que afectan la composición del aceite esencial de hojas de Pistacia lentiscus L. de València (España) Parâmetros do solo e características bioclimáticas que afetam a composição do óleo essencial das folhas de Pistacia lentiscus L. de Valência (Espanha)

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ABSTRACT

The variability of the soils found in an area together with the diversity of the bioclimatic parameters will affect the chemical profile of plant species, in our case *Pistacia lentiscus* L. The aim of this work is to analyse the bioclimatic characteristics and soil parameters affecting the essential oil (EO) composition of the leaves of the Pistacia lentiscus L. growing in València (Spain). The EO compositions of dried leaves of Pistacia lentiscus L. from five sampling sites with noticeable differences in soil and climate features were analysed by GC(MS) and GD(FID). The main bioclimatic and soil parameters were also determined in order to relate them to oil composition. α -pinene (2.8-39.2%), myrcene (0.6-59.3%), β -phellandrene (0.0-42.8%), germacrene-D (3.9-20.4%) and terpinen-4-ol (0.2-19.0%) were found to be the major compounds showing a high variability. The five sampling sites presented well-defined bioclimatic and edaphic characteristics that could be related to specific EO profiles. The results suggested that the EO composition of P. lentiscus L. depends more on the edaphic factors interacting with bioclimate conditions than on the geographical origin itself. Some general trends could be established based on the results: the Calcaric Arenosol (Saler) with a high evapotranspiration index was associated with a high sesquiterpenic fraction, (germacrene-D and β -caryophyllene, mainly). The Luvisol (Borrell and L'Ull), with high amounts of $\alpha + \beta$ -pinene, were found to be related to sub-humid bioclimatic conditions and clayey and fertile soils (high CEC and OM levels). This last requirement was also observed to be related to the myrcene content, but with drier climatic conditions and calcareous soils (Haplic Calcisol) with high level of carbonates and active lime (Lliria). The rest of hydrocarbon monoterpene and oxygenated monoterpene (terpinen-4-ol, mainly) fractions could be related to a dry climate and non-calcareous and low fertility soils (Eutric Regosol, Segart). Given the most suitable composition related to specific and useful biological activities, new research of controlled environmental factors (soil features and bioclimatic conditions) should be conducted in order to define the best conditions to manage industrial crops of P. lentiscus L.

RESUMEN

La variabilidad de los suelos que podemos encontrar en un área junto con la diversidad de los parámetros bioclimáticos afectará a la misma especie, en nuestro caso Pistacia lentiscus L., en su perfil químico. El objetivo de este trabajo es analizar

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las características bioclimáticas y los parámetros del suelo que afectan a la composición del aceite esencial (AE) de las bojas de la Pistacia lentiscus L. que crece en València (España). La composición de AE de las hojas secas de Pistacia lentiscus L, tomadas de cinco sitios de muestreo con diferencias notables en las características del suelo y el clima, se analizaron por GC(MS) y GD(FID). También se determinaron los principales parámetros bioclimáticos y edáficos para relacionarlos con la composición del aceite. α-Pineno (2,8-39,2%), mirceno (0,6-59,3%), β-felandreno (0,0-42,8%), germacreno-D (3,9-20,4%) y terpinen-4-ol (0,2-19,0%) fueron los compuestos principales que mostraron una alta variabilidad. Los cinco sitios de muestreo presentaron características bioclimáticas y edáficas bien definidas, que podrían estar relacionadas con perfiles específicos de AE. Los resultados obtenidos sugirieron que la composición de AE de P. lentiscus L. depende más de los factores edáficos que interactúan con las condiciones bioclimáticas que del propio origen geográfico. En base a los resultados se podrían establecer algunas tendencias generales: el Arenosol cálcico (Saler) con alto índice de evapotranspiración podría estar asociado con una alta tasa de fracción sesquiterpénica, (germacreno-D y β -cariofileno, principalmente). Se encontró que en los Luvisoles (Borrell y L'Ull), con altas cantidades de α + β -pineno, estaba relacionado con condiciones bioclimáticas subhúmedas y suelos arcillosos y fértiles (altos niveles de CIC y MO). Este último requerimiento también se observó relacionado con el contenido de mirceno, pero con condiciones climáticas más secas y suelos calcáreos (Calcisol háplico), con alto nivel de carbonatos y cal activa (Lliria). El resto de las fracciones de hidrocarburos monoterpenos y monoterpenos oxigenados (terpinen-4-ol, principalmente) podrían estar relacionados con suelos de clima seco y no calcáreos y de baja fertilidad (Regosol eútrico, Segart). Dada la composición más adecuada relacionada con actividades biológicas específicas y útiles, se deben realizar nuevas investigaciones de factores ambientales controlados (características del suelo y condiciones bioclimáticas) con el fin de definir las mejores condiciones para el manejo de cultivos industriales de P. lentiscus Τ.

RESUMO

A variabilidade dos solos que podemos encontrar numa área conjuntamente com a diversidade dos parâmetros bioclimáticos afetará o perfil químico duma espécie, no caso presente a Pistacia lentiscus L. O objetivo deste trabalho é analisar as características bioclimáticas e os parâmetros do solo que afetam a composição do óleo essencial (OE) das folhas de Pistacia lentiscus L. que cresce em Valência (Espanha). A composição OE das folhas secas de Pistacia lentiscus L., colhidas em cinco locais de amostragem com diferenças notórias no solo e nas características climáticas, foi analisada por GC-MS e GD-FID. Os principais parâmetros bioclimáticos e do solo também foram determinados e relacionados com a composição do óleo. Os principais compostos identificados e que mostraram alta variabilidade foram: α -pineno (2,8-39,2%), mirceno (0,6-59,3%), β-felandreno (0,0-42,8%), germacreno-D (3,9-20,4%) e terpineno-4-ol (0,2-19,0). Os cinco locais de amostragem apresentaram características bioclimáticas e edáficas bem definidas, que podem estar relacionadas com perfis específicos de OE. Os resultados obtidos sugerem que a composição do OE de P. lentiscus L. depende mais dos fatores edáficos que interagem com as condições bioclimáticas do que da própria origem geográfica. Com base nos resultados podem ser estabelecidas algumas tendências gerais: O Arenossolo cálcico (Saler) com elevado índice de evapotranspiração pode estar associado a uma alta taxa de fração sesquiterpénica, (principalmente germacreno-D e β-cariofileno). O Luvissol (Borrell e L'Ull), com elevadas quantidades de α + β -pineno, estava relacionado com condições bioclimáticas sub-búmidas e solos argilosos e férteis (altos níveis de CTC e MO). Observou-se também que este último requisito está relacionado com o teor de mirceno, mas com condições climáticas mais secas e solos calcários (Calcissolo Háplico), com alto teor de carbonatos e calcário ativo (Lliria). O restante das frações de hidrocarbonetos monoterpenos e monoterpenos oxigenados (principalmente terpineno-4-ol) podem estar relacionadas com clima seco e solos não calcários e de baixa fertilidade (Regossolo Êutrico, Segart). Dada a composição mais adequada estar relacionada com atividades biológicas específicas e úteis, devem realizarse novas investigações de fatores ambientais controlados (características do solo e condições bioclimáticas) para definir as melhores condições para a gestão de culturas industriais de P. Lentiscus L.

1. Introduction

Soil is a basic component for the development of plants and ecosystems. There is a partially bidirectional relationship between the vegetation and the characteristics of the soil; soils affect the vegetation and the vegetation itself conditions the soil properties. For this reason, the variability of the soils found in a given area will affect the proportion of both the inorganic and biochemical compounds of specific species. The bioclimatic variability that we find in a study area will also be another factor that will affect said composition.

KEYS WORDS

Volatile organic compounds, chemotypes, soilplant-bioclimatic relationships.

PALABRAS CLAVE

Compuestos orgánicos volátiles, quimiotipos, relaciones bioclimáticas sueloplanta.

PALAVRAS-CHAVE

Compostos orgânicos voláteis, quimiotipos, relações bioclimáticas soloplanta.

Pistacia lentiscus L. is a thermophilous dioecious evergreen shrub of the Anacardiaceae family growing wild in a wide range of habitats throughout the Mediterranean countries and the Canary Islands. It has been used in folk medicine as an antihypertensive, while the resin from its stems (gum mastic) is used for the treatment of digestive diseases (Vidrich et al. 2004) and dental hygiene. More recently, antimicrobial (El Idrissi. et al. 2016; Hafsé et al. 2013; Djenane et al. 2011; Haloui et al. 2015), insecticidal (Bougherra et al. 2015), herbicidal (Ismail et al. 2012), antifungal (Kordali et al. 2003; Barra et al. 2007), antioxidant (Barra et al. 2007; Bachrouch et al. 2015; Abdelkader et al. 2016) and other pharmacological activities have been reported (Aissi et al. 2016; Nahida et al. 2012; Catalani et al. 2017, Maaroufi et al. 2021).

The essential oil (EO) composition of its aerial parts (leaves and twigs, fruits and galls) has been studied extensively. Recent data from many studies carried out in different Mediterranean countries report the major components for each type of terpenic group (Llorens-Molina et al. 2015), showing an extensive chemical variability affecting both major compounds and total amounts of terpene types. This research has focused mainly on fresh or dried leaves and twigs.

Monoterpene hydrocarbons are considered as the predominant fraction in some studies. α and β -pinene, myrcene, limonene, β -phellandrene, sabinene and y-terpinene have been usually found as the major compounds (Boelens et al. 1991; Fernández et al. 2000; Zrira et al. 2003; Vidrich et al. 2004; Douissa et al. 2005; Dob et al. 2006; Gardeli et al. 2008; Amhamdi et al. 2009; Djenanne et al. 2011; Ismail et al. 2012; Hafsé et al. 2013; Haloui et al. 2015; Milia et al. 2020; Maaroufi et al. 2021). Samples rich in p-cymene (Barra et al. 2007), camphene (Mecheraraldjeri 2008) and δ -3-carene (Congiu et al. 2002; Haloui et al. 2015) have also been reported. Nevertheless, variability can be observed according to the harvest time and, above all, the place of sampling, even when closely spaced in the same country (Zrira et al. 2003; Vidrich et al. 2004; Dob et al. 2006; Gardeli et al. 2008; Mecherara-Idjeri 2008). Oxygen-containing monoterpenes in leaves shows also a great fluctuation from a very low level (Lo Presti et al. 2008) up to noticeable values (16.5%) (Milia et al. 2020), (36%) (Kivçak 2004), even higher than 50% (Zrira et al. 2003). Terpinen-4-ol is the most abundant and extended compound within this group. Relative high amounts of α and β -terpineol (Boelens et al. 1991; Dob et al. 2006) and bornyl acetate (Zrira et al. 2003) have been also cited. Regarding the sesquiterpenic (hydrocarbon and oxygenated) compounds, great differences have been described. As reported by Aissi et al. (2016), there are high amounts of this fraction (26.6-82.2%) in samples from Tunisia. Oils containing more than 80% of this fraction have been also cited by Lo Presti et al. (2008) while no more than 4% in other ones from Alghero (Sardinia) (Barra et al. 2007). Small amounts of diterpenes (dimyrcene isomers) have also been reported (Boelens et al. 1991; Lo Presti et al. 2008). This strong chemical variability agrees with the high degree of morphological and physiological plasticity related to its environmental tolerance and therefore, the wide range of habitats in which it grows (Nahum et al. 2008)).

Despite extensive knowledge regarding the EO composition of *P. lentiscus* L. leaves from a wide variety of locations, there are only data regarding the influence of pedologic and bioclimatic factors from Tunisia (Aissi et al. 2016). The aim of this work is to analyse the bioclimatic characteristics and soil parameters affecting the EO composition of leaves of *Pistacia lentiscus* L. found in València (Spain).

2. Materials and Methods

2.1. Sampling sites

Plant material and soil samples were collected from the same areas in autumn-winter 2016-2017; the coordinates and altitude of each one of the sampling zones are shown in **Table 1**. The most distant locations were approximately 100 km apart.

| Location | Coordinates | Altitude (m.a.s.l.) |
|--------------------------|-----------------------------|---------------------|
| Racó de Borrell (Gandia) | 38° 59' 04" N; 0° 16' 13" W | 208 |
| Font de L'Ull (Xeraco) | 39° 01' 19" N; 0° 15' 34" W | 175 |
| Les Mallades (Lliria) | 39° 37' 30" N; 0° 40' 46" W | 186 |
| El Saler (València) | 39° 22' 02" N; 0° 19' 33" W | 2 |
| Segart | 39° 40' 55" N; 0° 22' 33" W | 280 |

Table 1. Location of the sampling sites

2.2. Soil analyses and types

From each area, four soil samples of approximately 800 g each were taken at a depth of 0-20 cm and the distance between sampling points was around 500 m. Following the World Reference Base for Soil Resources (IUSS Working Group WRB 2015), the soils found in the five different areas were classified as follows: Chromic Luvisol (Racó de Borrell), Calcaric Luvisol (Font de L'UII), Calcaric Arenosol (El Saler), Haplic Calcisol (Les Mallades) and Eutric Regosol (Segart). Once the samples had evenly lost moisture, they were crushed with a roller to break aggregates and then passed through a 2 mm light sieve. Analyses were performed on fine soil (diameter < 2 mm). Soil texture was analysed by the hydrometer method (Bouyoucos 1962). Organic matter was analysed by the Walkey-Black method (Walkley and Black 1934). The carbonates were analysed using the Bernard Calcimeter (Loeppert and Suárez 1996). The active lime was analysed by the Drouineau method (Drouineau 1942). The CEC (cation exchange capacity) and exchangeable bases were analysed by Rhoades (Rhoades 1982). The following parameters were analysed in a saturation extract: pH, electrical conductivity (EC), chlorides, Na⁺, K⁺, Ca²⁺, and Mg²⁺. A Crison pH-meter Basic 20 and a Crison Conductimeter Basic 30 (HACH LANGE SPAIN, Barcelona, Spain) were used to measure pH and EC, respectively. Sodium and potassium were quantified with a PFP7 flame photometer (Jenway Inc., Burlington, VT, USA), chlorides were measured in a MKII Chloride Analyzer 926 (Sherwood, Inc., Cambridge, UK), and divalent cations ions of calcium and magnesium were measured with an atomic absorption spectrometer SpectrA 220 (Varian, Inc., CA, USA).

2.3. Climate data and bioclimatic indices

Climate data were obtained from the meteorological station nearest to each study area. The data covered a period of 17 years for five stations close to the sampling sites (Gandía, Tavernes de la Valldigna, Benifaió, Lliria and Moncada) and were collected from the SIAR net (SIAR 2018).

To investigate relationships between climatic characteristics, EO composition and soil parameters, several bioclimatic variables such as the Giacobbe index (Giacobbe 1938; Giacobbe 1959; Rivas-Martínez et al. 2011) were calculated. The bioclimatic parameters investigated are mostly related to summer, as this is a critical period for Mediterranean flora, and included: ETo, Penman-Monteith evapotranspiration; Ic, continentality index; It, thermicity index; Io, ombrothermic index; IG, summer drought index; PPv, summer precipitation for the three consecutive warmest months in the year; TPv, the sum of the monthly average temperature for the three consecutive warmest months (Table 2).

2.4. Plant material

Samples of approximately 200 g of fresh material of leaves and twigs were harvested from all around 20 individual shrubs randomly selected growing in five different locations (Table 1) (4 individuals x location). A voucher specimen was placed in the Herbarium of Mediterranean Agroforestry Institute at the Universitat Politècnica de València (Spain) (VALA 9581-9585). After removing damaged and yellowish material, each whole sample was dried in a dry and shady place at room temperature for 20 days just before EO extraction.

| Locality | lt ^a | lc ^b | loc | Ppv ^d | Ttve | ETof | Р | IG ^g |
|-----------------------|-----------------|-----------------|-----|------------------|-------|--------|-------|-----------------|
| Gandia (Borrell) | 387.8 | 15.5 | 2.7 | 37.0 | 740.9 | 1052.1 | 676.8 | 1.4 |
| Benifaio (Saler) | 390.3 | 15.2 | 1.8 | 38.2 | 738.0 | 1171.6 | 441.7 | 1.5 |
| Tavernes V (L'UII) | 412.0 | 14.2 | 2.3 | 61.0 | 740.1 | 1130.0 | 603.1 | 2.4 |
| Moncada (Segart) | 391.0 | 15.4 | 1.5 | 46.0 | 730.8 | 1125.1 | 370.9 | 1.8 |
| Lliria | 347.7 | 15.8 | 1.7 | 55.9 | 711.8 | 1101.3 | 394.3 | 2.3 |

Table 2. Bioclimatic indices of P. lentiscus L. corresponding to the sampling locations

T: yearly average temperature. m: average temperature of the minima of the coldest month of the year. M: average temperature of the maxima of the coldest month of the year. T max: average temperature of the warmest month. T min: average temperature of the coldest month. Tm: average temperature of each month. P: total yearly precipitation (mm). It^a: Thermicity index, 10 × (T + M + m). Ic^b: Continentality index, T max - T min. Io^c: Ombrothermic index, (P/12) × 10/ΣTm. Ppv^d: Summer precipitation in mm of the three consecutive warmest months in the year. Ttve: Value in tenths of a degree resulting from the sum of the monthly average temperatures of the three consecutive warmest months of the year. ETo^f: Evapotranspiration of Pemman-Monteith. IG^a (Giacobbe

index): (Pjun + Pjul + Paug)/Tm of the warmest month.

2.5. Essential oil extraction

After homogenizing each sample, 50 g was subjected to hydrodistillation for 3 h in a Clevenger-type apparatus. EO was swept away with 2 mL of dichloromethane (DCM). After removing the aqueous phase, the EO extract was dried over anhydrous sodium sulphate. The solvent was removed under reduced pressure, using a rotary evaporator Laborota 4001 (Heidolph Instruments, Schwabach, Germany) at 20 °C. Solutions containing 10 µL EO/1 mL of DCM were kept in the dark at -18 °C in sealed vials until analysis.

2.6. GC analysis

The analysis was carried out by gas chromatography with flame ionization detector (GC-FID) in a Clarus 500 GC (Perkin-Elmer Inc.) chromatograph equipped with a capillary column ZB-5 (30 m x 0.25 mm i.d. x 0.25 mm film thickness; Phenomenex Inc.). The injection volume was 1 μ L. The GC oven temperature was programmed from 50 °C to 250 °C at a rate of 3 °C min⁻¹. Helium was the carrier gas (1.2 mL min⁻¹). Detector temperature was set at 250 °C. The percentage composition of the EO was computed from GC peak areas without correction factors by means of the software

Total Chrom 6.2 (Perkin-Elmer Inc.). Analysis by GC-MS was carried out using a Clarus 500 GC-MS (Perkin-Elmer Inc.) equipped with the same column and programmed with the same oven temperatures mentioned above. Ionization source temperature was set at 200 °C and 70 eV electron impact mode was employed. MS spectra were obtained by means of total ion scan (TIC) mode (mass range m/z 45-500 uma). The total ion chromatograms and mass spectra were processed with the software Turbomass 5.4 (Perkin-Elmer Inc.). Kovats retention indices were determined by injection of C8-C25 nalkanes standard (Supelco®) under the same GC conditions. The essential oil components were identified by comparison of their mass spectra with those of computer library NIST MS Search 2.0 and available data in the literature (Adams 2007). The identification of the following compounds was confirmed by comparison of their experimental Kovats RI with those of authentic reference standards (Sigma-Aldrich®): α -pinene, β -pinene, camphene, myrcene, camphor, terpinolene, borneol, terpinen-4-ol, bornyl acetate and linalool.

2.7. Statistical analysis

The statistical analysis was carried out by means of one-way ANOVA, Principal Component

Analysis (PCA) and Discriminant Function Analysis, using Statgraphics XVII Centurion Software. As the raw data of EO composition were expressed as % peak areas, they were subjected to arcsine [square root (%/100)] transformation to reach data homoscedasticity. The average values were compared by Fisher's least significant difference (LSD) with 95% significance level.

3. Results

3.1. Results of soil parameters

Soil parameter analyses from the different sampled sites are displayed in **Table 3**. The five analysed soils show similarities and differences

between them. It should be noted that in relation to pH and electrical conductivity (EC), they have normal values when the types of soil are compared. The Saler soil has a slightly elevated EC value as it is a sandy soil of the dune zone near the sea. Calcareous soils such as Lliria, L'Ull and Saler have high values of carbonate and active lime. In general, the organic matter (OM) content is high at Lliria, L'Ull and Borrell. Adsorbed cations, cation exchange capacity (CEC) and exchangeable percentage of sodium (ESP) are normal, except in the Saler soil where the amount of Na and ESP is high, as expected in dune soils, with reduced fertility (CEC) and very low or no clay contents. In relation to the soil solution, it is also worth mentioning the high Cl⁻ and Na⁺ values.

As displayed in **Figure 1**, there were statistically significant differences (P < 0.05) between active lime, organic matter, cation exchange capacity,

| Location | Carbo (% | nates 5) | AL (‰) | OM (%) | Ca²⁺ (cmol _c /kg) | Mg² (cmol _c) (| ₂₊ /kg) (cı | Na⁺ nol _c /kg) | K⁺ (cmol _c /kg) | CEC (cmol _c /kg) | ESP (%) |
|----------|-------------|--------------|---------------------------|----------------|---------------------------------|-------------------------------|----------------|------------------------------|-------------------------------|--------------------------------|--------------------|
| Lliria | 45.2: | ±2.5 4 | 46.7±1.5 | 4.4±0.4 | 23.2±1.5 | 0.7±0 |).2 C | .1±0.0 | 0.7±0.1 | 27.2±6.1 | 0.3±0.1 |
| L'UII | 54.7: | ±2.6 4 | 45.9±2.1 | 4.1±1.7 | 15.3±3.0 | 1.1±0 |).3 C | .1±0.0 | 0.6±0.2 | 17.0±3.2 | 0.5±0.2 |
| Saler | 25.6: | ±2.8 | 6.3±0.5 | 1.4±0.8 | 8.8±2.0 | 0.7±0 |).1 C | .3±0.1 | 0.1±0.0 | 9.1±3.5 | 3.2±0.9 |
| Borrell | 0.1± | 0.1 | 1.2±0.8 | 4.3±2.0 | 12.0±4.1 | 1.1±0 |).3 C | .1±0.0 | 0.5±0.2 | 24.6±5.9 | 0.3±0.2 |
| Segart | 0.7± | 0.2 | 1.0±0.2 | 1.7±0.6 | 10.9±3.5 | 1.0±0 |).2 C | .1±0.0 | 0.5±0.2 | 13.0±3.0 | 0.9±0.2 |
| | | | Saturatio | n Extract | | | | Par | ticle size dist | ribution | |
| Location | рН | EC (dS/m) | CI [.] (mg/L) | Ca²+ (mg/L) | Na⁺ (mg/L) | Mg²+ (mg/L) | K⁺ (mg/L) | Sand (% | 5) Silt (%) | Clay (%) | Textural Class |
| Lliria | 7.5±0.2 | 0.6±0.1 | 26.3±3.7 | 48.5±9.0 | 12.1±3.1 | 6.6±1.3 | 10.6±2.3 | 57.9±12. | 5 34.4±15.6 | 7.6±3.6 | Sandy Ioam |
| L'UII | 7.8±0.2 | 0.7±0.2 | 26.5±2.9 | 43.3±9.9 | 19.7±4.5 | 10.5±1.6 | 33.8±4.5 | 58.0±21. | 9 32.5±21.2 | 9.5±1.9 | Sandy Ioam |
| Saler | 7.8±0.3 | 1.2±0.3 | 111.3±3.8 | 51.2±8.2 | 47.5±5.7 | 6.6±1.2 | 28.9±8.1 | 92.1±5.1 | 1 7.9±5.1 | 0.0±0.0 | Sand |
| Borrell | 7.5±0.1 | 0.4±0.0 | 48.0±6.7 | 21.8±2.5 | 25.0±5.1 | 6.5±1.6 | 5.2±1.6 | 63.5±12. | 9 6.6±1.8 | 29.9±11.3 | Sandy clay loam |
| Segart | 7.8±0.2 | 0.5±0.2 | 36.8±7.8 | 22.9±7.1 | 23.8±3.2 | 12.2±6.4 | 11.1±4.1 | 65.6±6. | 5 14.3±2.2 | 20.1±4.4 | Sandy clay loam |

Table 3. Soil parameters from the different sampling sites

AL: Active Lime; OM: Organic Matter; CEC: Cation Exchange Capacity; ESP: Exchange Sodium Percentage; EC: Electrical Conductivity.

[SOIL PARAMETERS AND BIOCLIMATIC CHARACTERISTICS AFFECTING ESSENTIAL OIL COMPOSITION OF LEAVES OF PISTACIA LENTISCUS L. FROM VALÈNCIA (SPAIN)]



Figure 1. Mean values and LSD Fisher intervals (95%) of active lime, organic matter, cation exchange capacity, exchangeable sodium percentage, sand, silt and clay of soil at different sampling sites where the *Pistacia lenticus* L- leaves were sampled. Differences among mean values with different letters are statistically significant (P < 0.05).

exchangeable sodium percentage, sand, silt and clay when the one-way ANOVA was applied. High individual variability means among the different within sampling site could be observed in every one of the analysed properties (Table 3). The most relevant ones were submitted to ANOVA analysis (Figure 1). Active lime (AL) showed statistically significant differences (P < 0.001), with the higher values in Lliria and L'UII. With regard to the OM level, two groups of sampling sites were found: Saler and Segart accounted for a significantly lower level (P < 0.01) with respect to Borrell, Lliria and L'Ull. The Cation Exchange Capacity (CEC) showed high variability, with Borrell, Lliria and L'Ull being significantly more fertile. In the same way, EC of saturation extracts presented statistically significant differences (P < 0.001). The Saler value was significantly higher than the other ones.

As for sand, silt and clay, significant differences were also found (P < 0.01, P < 0.01, P < 0.01, respectively). Obviously, the highest level of sand (and the lowest level of clay) was found in Saler (coastal site). By contrast, Borrell and Segart were found to be the richest in clay, whereas Lliria and L'UII had a higher silt content. A clear dichotomy was found between Saler and the rest of sampling points related to ESP, which showed a significantly higher content.

3.2. Essential oil composition

The essential oil composition of the 20 individuals sampled in the studied sites is displayed in **Table 4**. The major compounds were found to be similar to the ones reported in the literature, but great individual differences were noted even within the same location. Of the total compounds, hydrocarbon monoterpenes accounted for between 35.8% (El Saler, M1) and 74.9% (Segart, M1), oxygenated monoterpenes from 0.2% (Segart, M1) to 22.5% (El Saler, M1) and hydrocarbon sesquiterpenes from 12.3% (L'UII, M4) to 43.9% (Lliria, M1). Oxygenated sesquiterpene and diterpene fractions accounted for no more than 3.1% (El Saler, M1) and 0.8% (Lliria, M4), respectively.

As for major compounds, α -pinene (2.8-39.2%), myrcene (0.6-59.3%) and β -phellandrene (0.0-42.8%), germacrene-D (3.9-20.4%) and terpinen-4-ol (0.2-19.0%) reached the highest individual levels. Noteworthy amounts of β -caryophyllene (3.4-17.1%), β -pinene (0.3-17.1%), δ -3-carene (0.2-15.7%), sabinene (0.0-15.3%), limonene (0.1-6.1%) and γ -terpinene (0.0-6.0%) were also detected.

Despite the great individual variability, some statistically significant differences (P < 0.05) were found when one-way ANOVA was applied in order to compare the amounts of major and grouped compounds according to the individual's sampling points (Table 5). The hydrocarbon monoterpene fraction, as a whole, was significantly lower in Saler (and Lliria, without achieving statistical significance). Within this fraction, the amount of myrcene showed a different tendency, with a significant increase in Lliria and Segart (this last one with no significant differences). The content of α -pinene was significantly higher in Borrell and L'Ull compared to Lliria and Segart (β-pinene also showed a similar variation). Likewise, limonene was significantly lower in Lliria and Segart with respect to the rest of sampling sites, whereas β -phellandrene was not found in Lliria. The percentage of terpinen-4-ol was significantly higher in Saler with respect to Borrell. As for the sesquiterpenic fraction, Lliria and Saler presented a significant increase if compared to the rest of locations.

[SOIL PARAMETERS AND BIOCLIMATIC CHARACTERISTICS AFFECTING ESSENTIAL OIL COMPOSITION OF LEAVES OF PISTACIA LENTISCUS L. FROM VALÈNCIA (SPAIN)]

Table 4. Essential oil composition for the 20 samples

| Compounds ¹ | RI ² | Id. Met. ³ | | BOR | RELL | | | LLI | RIA | | | Ľ | UII | | | EL S | ALER | | | SEG | ART | |
|------------------------------------|------------------------|-----------------------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|
| | | | M1 | M2 | M3 | M4 | M1 | M2 | M3 | M4 | M1 | M2 | M3 | M4 | M1 | M2 | М3 | M4 | M1 | M2 | M3 | M4 |
| Tricyclene | 925 | MS, RI | 0.6 | 0.4 | 0.1 | _4 | 0.3 | 0.1 | - | tr⁵ | - | 0.3 | 0.1 | 0.5 | 0.1 | 0.3 | 0.5 | 0.5 | tr | - | - | 0.2 |
| α-Thujene | 929 | MS, RI, St. | 1.9 | 0.2 | 0.3 | 0.1 | 0.1 | tr | - | tr | 0.2 | 0.1 | tr | 0.3 | tr | 0.3 | 0.1 | 0.1 | tr | 0.1 | 0.3 | - |
| α-Pinene | 937 | MS, RI | 15.2 | 23.7 | 32.6 | 31.6 | 12.2 | 4.1 | 24.8 | 4.2 | 20.5 | 11.9 | 39.2 | 25.7 | 6.9 | 15.1 | 13.3 | 13.5 | 2.8 | 3.3 | 13.6 | 12.4 |
| Camphene | 953 | MS, RI, St. | 2.7 | 2.2 | 0.3 | 0.4 | 1.6 | 0.4 | 0.3 | 0.2 | 0.4 | 1.6 | 0.5 | 2.1 | 0.8 | 1.4 | 2.6 | 2.6 | tr | 0.1 | 0.1 | 0.2 |
| Sabinene | 975 | MS, RI | 8.2 | 3.2 | 9.3 | 5.2 | 8.3 | 0.8 | 0.2 | 0.6 | tr | 4.2 | 1.0 | 15.3 | 4.5 | 8.9 | 5.1 | 2.6 | tr | 3.4 | - | 11.1 |
| β-Pinene | 979 | MS, RI, St. | 5.6 | 10.1 | 10.4 | 10.6 | 5.6 | 0.7 | 7.4 | 0.5 | 16.3 | 2.9 | 17.1 | 2.1 | 1.0 | 1.4 | 6.5 | 6.2 | 0.3 | 0.7 | 8.2 | 0.4 |
| Myrcene | 996 | MS, RI, St. | 1.0 | 1.0 | 0.8 | 0.6 | 1.4 | 59 | 2.1 | 54.7 | 0.8 | 0.8 | 0.7 | 0.8 | 0.9 | 1.4 | 0.8 | 1.3 | 59.3 | 3.2 | 1.5 | 1.3 |
| δ-3-Carene | 1010 | MS, RI | 1.1 | 12.4 | 0.7 | 8.8 | 0.3 | 0.2 | 10.1 | 4 | 8.2 | 7.8 | 1.3 | 0.7 | 2.2 | 5.4 | 8.4 | 7.2 | 6.9 | 4.5 | 15.7 | 8.8 |
| α-Terpinene | 1019 | MS, RI, St. | 3.7 | 1.9 | 2.3 | 1.8 | 1.5 | 0.5 | 0.3 | 0.3 | 2.8 | 2.2 | 0.5 | 3.6 | 1.8 | 3.7 | 1.5 | 1.9 | 0.2 | 1.6 | 2.1 | 3.1 |
| p-Cymene | 1027 | MS, RI, St. | 0.7 | 1.3 | 0.4 | - | - | - | 0.7 | 0.2 | 1.5 | 2.6 | 0.2 | 1.4 | - | 0.7 | 0.4 | 0.4 | 0.3 | - | - | - |
| Limonene | 1032 | MS, RI, St. | 6.1 | 4.9 | 2.7 | 1.5 | 0.4 | 0.1 | 3.8 | 1.2 | 3.3 | 4.6 | 2.9 | 1.5 | 5.3 | 2.8 | 3.4 | 2.7 | 1.9 | 0.5 | 1.6 | 2.3 |
| b-Phellandrene | 1034 | MS, RI, | 3.1 | 8.5 | 4.3 | 12.6 | - | - | - | - | 7.2 | 4.3 | 5.9 | 2.6 | 6.5 | 3.9 | 5 | 5.5 | 2.4 | 42.8 | 12.6 | 8.7 |
| (Z)-β-Ocimene | 1040 | MS, RI | - | - | - | - | 2.5 | 0.3 | 4.9 | 1.9 | - | - | - | - | - | - | - | - | - | - | - | - |
| (E)-β-Ocimene | 1052 | MS, RI | - | - | - | - | 0.2 | - | 0.3 | 0.3 | - | tr | - | 0.3 | - | - | - | - | 0.6 | - | 0.5 | 0.2 |
| γ-Terpinene | 1061 | MS, RI, St. | 5.8 | 2.4 | 3.7 | 2.2 | 3.3 | 1.3 | 1.2 | 1.2 | 4.1 | 4.1 | 1.2 | 5.9 | 4.5 | 6 | 2.2 | 3.5 | - | - | - | - |
| Terpinolene | 1086 | MS, RI, St. | - | - | - | - | - | - | - | - | tr | - | - | - | - | - | - | - | 0.2 | 2.6 | 3.3 | 4.7 |
| p-Cymenene | 1090 | MS, RI | 1.8 | - | 1.4 | 1.0 | 0.7 | 0.3 | 0.4 | 0.4 | 1.0 | 1.2 | 1.7 | 1.7 | 1.4 | 1.6 | 1.3 | 1.4 | - | 1.4 | 1.0 | 1.4 |
| H. monoterpenes | | | 57.3 | 72.1 | 69 | 76.3 | 38.5 | 67.6 | 56.6 | 69.9 | 66.4 | 48.6 | 72.2 | 64.3 | 35.8 | 52.9 | 51 | 49.4 | 74.9 | 64.2 | 60.6 | 54.6 |
| Linalool | 1097 | MS, RI, St. | - | - | 0.4 | - | 0.5 | tr | 0.3 | 0.2 | - | 0.1 | tr | 0.1 | 0.3 | - | - | - | - | tr | tr | - |
| Mentha-2-en-1-ol <z-p></z-p> | 1125 | MS, RI | - | tr | tr | - | - | - | tr | - | - | - | tr | - | - | - | - | - | - | - | - | - |
| 3-Terpinen-1-ol | 1134 | MS, RI | tr | - | 0.1 | tr | 0.1 | - | tr | - | - | 0.1 | tr | 0.1 | 0.4 | 0.1 | tr | tr | - | tr | 0.1 | 0.2 |
| Mentha-2-en-1-ol < <i>E</i> -p> | 1141 | MS, RI | - | - | - | tr | - | - | - | - | tr | - | - | - | - | - | - | - | - | - | - | - |
| Camphor | 1148 | MS, RI, St. | - | - | - | - | - | tr | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| (Z)-β-terpineol | 1152 | MS, RI | - | - | - | tr | tr | - | - | - | - | tr | - | 0.1 | 0.2 | tr | tr | - | - | - | tr | 0.1 |
| Terpinen-4-ol | 1181 | MS, RI, St. | 15.4 | 5.3 | 7.4 | 3.5 | 7.1 | 1.8 | 0.5 | 0.7 | 9.4 | 9.1 | 1.4 | 12.9 | 19 | 13.1 | 4.0 | 5.8 | 0.2 | 5.6 | 8.6 | 14.8 |
| α-Terpineol | 1194 | MS, RI | - | - | - | - | tr | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| Verbenone | 1204 | MS, RI, St. | 1.9 | - | 1.3 | - | 0.5 | 0.8 | 1.5 | 0.8 | - | 0.4 | 2.1 | 2.1 | 2.6 | - | - | - | - | - | - | 1.4 |
| (E)-Piperitol | 1205 | MS, RI | - | 0.6 | - | - | - | - | - | - | 0.1 | - | - | - | - | 0.1 | 0.1 | 0.1 | - | 0.1 | 0.3 | - |
| β-Cyclocitral | 1223 | MS, RI | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | tr | - |
| Bornyl acetate | 1291 | MS, RI, St. | 0.2 | - | - | - | - | - | tr | - | - | - | tr | - | tr | - | - | - | - | - | - | - |
| Carvacrol | 1307 | MS, RI, St. | 0.2 | - | 1.3 | - | - | - | 0.4 | - | - | - | 0.5 | - | - | - | - | - | - | - | - | - |
| Ox. monoterpenes | | | 17.7 | 5.9 | 10.3 | 3.5 | 8.2 | 2.6 | 2.8 | 1.7 | 9.5 | 9.7 | 4.1 | 15.2 | 22.5 | 13.3 | 4.1 | 5.8 | 0.2 | 5.8 | 8.9 | 16.5 |

Table 4. Essential oil composition for the 20 samples (cont.)

| Compounds ¹ | RI ² | Id. Met. ³ | | BOR | RELL | | | LL | RIA | | | Ľ | UII | | | EL S | ALER | | | SEG | ART | |
|--|------------------------|-----------------------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|------|
| | | | M1 | M2 | M3 | M4 | M1 | M2 | M3 | M4 | M1 | M2 | М3 | M4 | M1 | M2 | M3 | M4 | M1 | M2 | M3 | M4 |
| δ-Elemene | 1338 | MS, RI | 0.3 | - | 0.6 | - | - | 0.8 | - | - | - | - | 0.7 | - | 0.2 | 0.5 | 1.1 | 0.9 | - | 0.5 | tr | - |
| α-Copaene | 1373 | MS, RI | - | tr | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | tr | - | - |
| β-Bourbonene | 1380 | MS, RI | - | - | - | - | 0.1 | tr | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| β-Elemene | 1391 | MS, RI | 0.4 | tr | - | - | 1.1 | 0.6 | 0.1 | 0.1 | 0.1 | 0.8 | 0.1 | 0.2 | - | tr | tr | tr | 0.5 | - | tr | tr |
| β-Caryophyllene | 1415 | MS, RI | 8.7 | 12.4 | 9.7 | 9.3 | 15.3 | 11.7 | 14.5 | 9.6 | 11.4 | 6.6 | 7.9 | 3.4 | 13.6 | 11.7 | 14.7 | 17.1 | 3.5 | 11.7 | 14.4 | 4.8 |
| β-Copaene | 1432 | MS, RI | 0.1 | 0.1 | 0.1 | - | 0.1 | tr | 0.1 | 0.1 | tr | 0.2 | - | tr | - | 0.1 | 0.1 | 0.2 | 0.1 | tr | 0.1 | 0.1 |
| (Z)-Muurola-3,5-diene | 1450 | MS, RI | - | - | - | - | - | - | 0.4 | tr | - | - | - | - | - | - | - | - | tr | - | - | tr |
| α-Humulene | 1451 | MS, RI | 0.1 | - | tr | - | 0.1 | 0.1 | 0.1 | tr | - | - | - | 0.1 | tr | 0.1 | tr | 0.1 | 0.1 | 0.1 | tr | tr |
| Aromadendrene <allo></allo> | 1456 | MS, RI | 1.2 | 0.9 | 1.0 | 0.7 | 2.4 | 1.1 | 1.9 | 1.3 | - | 0.2 | 0.1 | 0.2 | 2.1 | 1.6 | 1.6 | 1.7 | 1.0 | 1.6 | 1.2 | 0.4 |
| γ-Gurgujene | 1472 | MS, RI | 1.1 | - | - | - | tr | tr | 1.1 | 0.1 | 0.7 | 1.8 | 0.9 | 0.8 | 0.1 | 0.1 | 0.1 | tr | 0.2 | 0.1 | tr | 0.1 |
| Germacrene-D | 1485 | MS, RI | 6.4 | 6.0 | 6.1 | 5.3 | 20.4 | 9 | 13.5 | 9.4 | 9.3 | 14 | 8.7 | 3.9 | 7.9 | 13 | 17.3 | 15.2 | 11.5 | 9.7 | 5.3 | 11 |
| Bicyclogermacrene | 1490 | MS, RI | tr | - | 0.1 | - | 0.1 | 0.1 | 0.1 | 0.1 | - | - | 0.1 | - | tr | tr | 0.1 | 0.1 | tr | 0.1 | tr | 0.1 |
| (<i>E</i>)-Muurola-4(14),5- diene | 1494 | MS, RI | 0.1 | - | 0.1 | - | 0.5 | 0.2 | 0.3 | tr | - | 0.4 | 0.9 | 0.2 | tr | tr | 0.2 | 0.3 | 0.2 | - | 0.2 | 0.3 |
| α-Muurolene | 1500 | MS, RI | 0.2 | - | 0.2 | 0.1 | 1.7 | 0.6 | 1.1 | 0.4 | tr | 0.3 | 0.2 | 0.2 | 0.2 | 0.3 | 0.4 | 0.3 | 0.4 | 0.2 | 1.8 | 1.2 |
| β-Cadinene | 1501 | MS, RI | 0.6 | 0.2 | 0.2 | 0.4 | - | - | - | - | - | - | - | - | - | - | - | - | 0.5 | 0.4 | 0.3 | 0.5 |
| β-Bisabolene | 1504 | MS, RI | tr | 0.1 | 0.1 | - | 0.9 | 0.4 | 0.5 | 0.4 | 0.1 | 1 | 0.4 | 0.6 | 0.8 | 0.5 | 0.4 | 0.5 | - | - | - | - |
| (<i>E,E</i>)-α-Farnesene | 1506 | MS, RI | - | - | - | - | tr | 0.3 | 0.1 | tr | - | 0.1 | 0.2 | - | - | 0.1 | 0.3 | 0.4 | 0.1 | 0.2 | tr | tr |
| γ-Cadinene | 1508 | MS, RI | 0.1 | - | 0.1 | 0.1 | 0.4 | 0.2 | 0.6 | 0.2 | - | 0.1 | 0.1 | 0.1 | 0.2 | 0.2 | 0.2 | 0.2 | 0.1 | 0.1 | 0.2 | 0.6 |
| δ-Cadinene | 1514 | MS, RI | 0.4 | 0.3 | 0.2 | 0.1 | 0.5 | 0.6 | 0.5 | 0.4 | - | - | tr | - | 0.1 | - | 0.2 | - | 0.3 | 0.3 | 0.3 | 0.3 |
| (E)-Cadina-1,4-diene | 1528 | MS, RI | 2.2 | 0.7 | 0.6 | 1.7 | 0.3 | 1.3 | 2.3 | 1.9 | 0.5 | 3.7 | 1.2 | 2.4 | 2.5 | 1.7 | 1.4 | 1.6 | 2.2 | 1.5 | 1.3 | 2.0 |
| Zonarene | 1530 | MS, RI | 0.2 | - | - | - | 0.2 | - | 0.1 | 0.1 | - | 0.6 | 0.1 | 0.3 | 0.4 | tr | 0.1 | - | 0.2 | 0.1 | - | 0.1 |
| (<i>E</i>)-γ-Bisabolene | 1531 | MS, RI | - | - | - | - | - | - | - | - | - | - | tr | - | - | - | 0.1 | - | - | - | - | - |
| α-Cadinene | 1533 | MS, RI | - | - | - | - | - | - | 0.1 | - | - | 0.2 | 0.1 | - | - | - | 0.1 | tr | - | - | - | - |
| H. sesquiterpenes | | | 22.0 | 20.6 | 18.9 | 17.9 | 43.9 | 26.9 | 37.2 | 24.1 | 22.1 | 29.9 | 21.5 | 12.3 | 27.9 | 29.9 | 38.1 | 38.6 | 21.0 | 26.5 | 25.1 | 21.5 |
| Elemol | 1550 | MS, RI | - | - | - | - | - | tr | - | - | - | tr | tr | - | - | - | 0.1 | tr | - | - | - | - |
| (E)-Nerolidol | 1563 | MS, RI | 0.2 | - | 0.4 | - | - | 0.5 | 0.1 | - | tr | - | - | 0.3 | 0.3 | 0.3 | 0.8 | 0.7 | - | 0.4 | - | - |
| Globulol | 1585 | MS, RI | - | - | - | - | 0.1 | - | - | - | - | - | - | - | 1.7 | tr | - | - | - | - | - | 0.2 |
| Viridiflorol | 1593 | MS, RI | - | tr | 0.1 | - | 0.1 | - | - | tr | - | 0.2 | tr | 0.1 | tr | - | - | - | - | 0.1 | 0.2 | 0.1 |
| Rosifoliol | 1599 | MS, RI | - | - | - | - | tr | - | - | - | - | tr | - | - | tr | - | 0.1 | tr | - | - | tr | 0.2 |
| Junenol | 1605 | MS, RI | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | tr | - | tr |
| Cubenol <1,10-di-epi> | 1622 | MS, RI | - | - | - | - | - | - | - | tr | - | tr | - | - | 0.2 | - | - | - | - | - | - | - |
| γ-Eudesmol | 1632 | MS, RI | - | tr | tr | - | tr | - | tr | - | - | 0.2 | 0.1 | 0.1 | 0.2 | tr | 0.1 | 0.1 | tr | 0.1 | - | 0.1 |



[SOIL PARAMETERS AND BIOCLIMATIC CHARACTERISTICS AFFECTING ESSENTIAL OIL COMPOSITION OF LEAVES OF PISTACIA LENTISCUS L. FROM VALÈNCIA (SPAIN)]

Table 4. Essential oil composition for the 20 samples (cont.)

| Compounds ¹ | RI ² | ld. Met. ³ | | BOR | RELL | | | LLI | RIA | | | Ľ | UII | | | EL S/ | ALER | | | SEG | ART | |
|-------------------------------|------------------------|-----------------------|------|------|------|------|------|------|------|------|------|------|------|------|------|-------|------|------|------|------|------|------|
| Cadinol-epi-α | 1638 | MS, RI | 0.3 | - | tr | - | 0.2 | tr | 0.1 | 0.1 | - | 0.9 | 0.1 | 0.4 | 0.6 | 0.1 | 0.2 | 0.2 | 0.2 | 0.2 | - | 0.1 |
| Muurolol-epi-α | 1640 | MS, RI | 0.1 | tr | _ | - | - | - | _ | _ | - | - | - | 0.5 | 0.1 | - | tr | - | - | - | - | - |
| α-Muurolol | 1642 | MS, RI | - | - | tr | - | - | - | 0.1 | _ | 0.4 | - | - | - | - | - | 0.1 | tr | - | - | - | tr |
| Ox. sesquiterpenes | | | 0.6 | - | 0.5 | - | 0.5 | 0.5 | 0.3 | 0.1 | 0.5 | 1.3 | 0.2 | 1.3 | 3.1 | 0.5 | 1.2 | 1.0 | 0.2 | 0.7 | 0.2 | 0.7 |
| Dihydromyrcene (isomer) | 1949 | MS, RI | 0.1 | tr | 0.1 | - | tr | tr | - | 0.6 | - | tr | - | - | - | - | tr | tr | tr | - | - | tr |
| Manoyl oxide | 1991 | MS, RI | - | tr | tr | - | - | - | - | 0.2 | - | - | - | - | - | - | - | - | - | - | - | tr |
| Diterpenoids | | | 0.1 | - | 0.1 | - | - | - | - | 0.8 | - | - | - | - | - | - | tr | tr | tr | - | - | - |
| 2-Hexenal | 855 | MS, RI | - | - | - | - | tr | tr | - | tr | - | - | - | - | - | - | - | - | - | - | - | - |
| 2-Nonanone | 1092 | MS, RI | - | 0.1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 0.1 | - | - | - |
| Nonanal | 1105 | MS, RI | tr | - | - | - | tr | - | 0.1 | 0.1 | - | tr | 0.1 | - | 0.3 | 0.3 | tr | - | - | - | - | - |
| Isopentyl isovalerate | 1107 | MS, RI | 0.1 | - | 0.1 | - | - | - | - | - | 0.1 | - | - | - | - | - | - | - | - | 0.1 | - | - |
| 2-Methyl butyl hexa- noate | 1253 | MS, RI | - | - | - | - | - | - | - | - | - | - | 0.1 | - | - | - | - | - | 0.1 | - | - | - |
| 2-Undecanone | 1292 | MS, RI | - | - | - | - | 0.9 | - | - | - | 0.2 | 1.3 | 0.4 | 0.6 | _ | - | 0.3 | 0.3 | - | - | 0.1 | 0.1 |
| (Z)-3-Hexenyl n- hexanoate | 1379 | MS, RI | 0.5 | tr | tr | - | 0.4 | 0.1 | 0.4 | 0.1 | - | 0.9 | 0.1 | 0.5 | 0.5 | 0.4 | 0.1 | 0.1 | 0.6 | 0.2 | 0.1 | 0.1 |
| 3-Methyl-1-butyl benzoate | 1441 | MS, RI | - | - | - | - | - | - | tr | - | - | - | - | - | - | - | - | - | tr | - | - | - |
| Other | | | 0.6 | 0.2 | 0.1 | 0.0 | 1.4 | 0.1 | 0.5 | 0.2 | 0.4 | 2.2 | 0.7 | 1.1 | 0.7 | 0.6 | 0.4 | 0.4 | 0.8 | 0.2 | 0.2 | 0.3 |
| Total identified | | | 98.2 | 98.8 | 99.0 | 97.7 | 92.5 | 97.8 | 97.3 | 96.8 | 98.8 | 91.6 | 98.7 | 94.3 | 90.2 | 97.3 | 95 | 95.5 | 97.2 | 97.6 | 95.1 | 93.7 |

¹Constituents listed in order of increasing retention indices (RI) for each group of compounds. Unidentified components accounting for less than 0.5% are not reported. ²Kovats retention indices referred to n -alkanes, determined on a DB5 capillary column. ³Method of identification: all the reported compounds have been identified by comparison of GC-MS data of NIST computer mass library and Kovats RI with those reported by R. P. Adams (35). α-pinene, β- pinene, myrcene, α-terpinene, p-cymene, limonene, terpinolene, (Z)-thujone, (E)-thujone, camphor and terpinen-4-ol were identified by comparison of RI with those of authentic samples. ⁴Not detected. ⁵Percentage values less than 0.1% are denoted as tr (traces).



| Compounds | Borrell | Lliria | L'UII | El Saler | Segart |
|----------------------------|-------------------------|---------------------------|----------------|---------------|--------------------------|
| α -Pinene | 25.7 ± 8.1 a | 11.3 ± 9.8 b | 24.3 ± 11.4 ac | 12.2 ± 3.6 bc | 8 ± 5.7 b |
| Sabinene | 6.4 ± 2.8 a | 2.5 ± 3.9 a | 5.1 ± 7.0 a | 5.3 ± 2.7 a | 3.6 ± 5.2 a |
| β-Pinene | 9.1 ± 2.4 a | 3.6 ± 3.5 ab | 9.6 ± 8.2 a | 3.8 ± 3 ab | 2.4 ± 3.9 b |
| Myrcene | $0.8 \pm 0.2 \text{ b}$ | 29.3 ± 31.9 a | 0.8 ± 0.1 b | 1.1 ± 0.3 b | 16.3 ± 28.7 ab |
| δ-3-Carene | 5.7 ± 5.8 a | 3.7 ± 4.7 a | 4.5 ± 4.1 a | 5.8 ± 2.7 a | 9.0 ± 4.8 a |
| Limonene | 3.8 ± 2.1 a | 1.4 ± 1.7 b | 3.1 ± 1.3 ab | 3.5 ± 1.2 a | 1.6 ± 0.8 b |
| β-Phellandrene | 7.1 ± 4.3 a | 0 ± 0 b | 5.0 ± 2.0 a | 5.2 ± 1.1 a | 16.6 ± 17.9 a |
| Terpinen-4-ol | 7.9 ± 5.3 ab | $2.5 \pm 3.1 \text{ b}$ | 8.2 ± 4.8 ab | 10.5 ± 6.9 a | 7.3 ± 6.1 ab |
| β-Caryophyllene | 10 ± 1.6 abc | $12.8 \pm 2.6 \text{ ab}$ | 7.3 ± 3.3 c | 14.3 ± 2.3 a | $8.6 \pm 5.3 \text{ bc}$ |
| Germacrene-D | 5.9 ± 0.5 b | 13.0 ± 5.3 a | 9.0 ± 4.1 ab | 13.3 ± 4.0 a | 9.4 ± 2.8 ab |
| Hydrocarbon monoterpenes | 68.7 ± 8.2 a | 58.2 ± 14.3 ab | 62.9 ± 10.1 a | 47.3 ± 7.8 b | 63.6 ± 8.5 a |
| Oxygenated monoterpenes | 9.4 ± 6.3 a | 3.8 ± 3 a | 9.6 ± 4.5 a | 11.5 ± 8.4 a | 7.9 ± 6.8 a |
| Hydrocarbon sesquiterpenes | 19.9 ± 1.8 a | 33.0 ± 9.2 b | 21.4 ± 7.2 a | 33.7 ± 5.5 b | 23.6 ± 2.7 a |

Table 5. Differences among EO chemical profiles coming from the studied locations,concerning main and grouped compounds (Mean values, 0.5 SD and differences (P < 0.05)</td>LSD intervals are indicated by means of different letters on the same row)

4. Discussion

In order to define the distinctive edaphic and bioclimatic features of the five sampling locations affecting EO composition, a discriminant analysis based on the main soil and climate parameters and the main oil compounds was performed (**Figure 2**). Three functions with P < 0.05 were defined with λ Wilks values: 2.9869 E-7, 7.4627 E-5 and 7.386 E-3, respectively. A first distinction can be pointed out according to the content of active lime. Soils of calcareous nature were found in three sampling sites (Saler, L'Ull, Lliria), while those of Segart and Borrell have lower carbonate contents.

Within each of these groups, axis 1 marks an increasing fertility owing to the higher levels of active lime, clay and CEC (cation exchange capacity). On the other hand, it is worth mentioning the salinity observed in Saler soils, evidenced by its conductivity value, as well as the high chloride and sodium content because of its proximity to the sea. From the bioclimatic point of view, axis 1 also marks the difference between three of the locations Segart and Saler), drier than Lliria, whereas axis 2 separates L'Ull and Borrell by the increment in organic matter, clay contents and the subhumid climate. Further

interpretation connecting this trend with the oil composition was obtained by Principal Component Analysis (PCA) with compounds defining the main chemotypes of *P. lentiscus* L. EO reported in the literature. These results are graphically represented for clarity (Figure 3) and subjected to discussion.

The obtained plot according to axes 1 (24.9% of the variance) and 2 (21.0% of the variance) grouped the samples in such a way that more clear linkages between bioclimatic and soil parameters and EO chemical profiles were observed. Thus, Borrell, with the highest fertility and subhumid conditions was characterized by the pinene chemotype, whereas Lliria, with a higher rate of active lime, lower clay content and drier climate was characterized by the high proportion of myrcene, although it should be taken into account that the levels of this compound have shown great variability between samples, both in Lliria and in Segart.

Likewise, the Giacobbe index leads to the consideration that this location is susceptible to water stress. In this respect, it is worth mentioning the reported data from Aissi et al. (2016), in which the higher levels of myrcene were found in upper semi-arid climates and calcareous soils.

The hydrocarbon sesquiterpene fraction (germacrene-D and β -caryophyllene, mainly) was the major one in Saler samples. The edaphic and bioclimatic characteristics of this location were remarkably specific, as evidenced by a strong conductivity and a high value of evapotranspiration (ETo) and Exchangeable Sodium Percentage (ESP), together with a low proportion of active lime and no occurrence of clay. In this case, these data do not agree with those reported by Aissi et al. (2016) in which the samples rich in the sesquiterpene fraction come to be associated with sub-humid and lower humid climates. In this sense, it is necessary to indicate that the most similar locations to the last ones were Borrell and L'Ull, with a high ombrothermal index. Contrary to what is reported in Aissi et al. (2016), α + β -pinene and myrcene were the predominant compounds instead of the sesquiterpene fraction. Since in this work, the detailed soil parameters are not considered, this fact leads to the suggestion that the major influence of soil fertility is a consequence of the high rates of CEC, OM and clay.

Samples from Segart were rich in the monoterpenic fraction other than $\alpha + \beta$ -pinene and terpinen-4-ol. This location was characterized by

its lower ombrothermal index than Borrell and L'Ull, related to a lower average annual rainfall and a high summer drought. Average values of terpinen-4-ol were also found in a moderately high proportion (2.5-10.5%). These amounts were similar to those reported by Llorens-Molina et al. (2015), in which two sites coinciding with those studied in this work (Segart and L'Ull) were 5.1-6.7% compared to 7.2-8.3%, respectively. No significant differences were found between these sites for this compound in both studies. These values also agree with those referred by Aissi et al. (2016) (2.7-9.6%), with the higher values in upper semi-arid locations.

On the other hand, with respect to Saler, it also showed a major influence of evapotranspiration and EC. It is worth mentioning the specific features of sample 4 from Segart, which can be considered as a transition between siliceous and calcareous soils. Sample 4 has a greater content of active lime than the other three and is slightly more fertile (higher values of EC (0.8 dS/m) and has more organic matter (OM) (2.2%). According to the aforementioned tendency, it also showed a higher myrcene content than the Lliria and Segart oils.



Figure 2. Discriminant Analysis: soils, climatic characteristics and oils. Discriminant functions (standardized coefficients):



Figure 3. Biplot of Principal Components Analysis: EO composition-soils-climatic characteristics.

 $\label{eq:component 2: } 0.0585814^*ESP + 0.204888^*CE SE + 0.323235^*Active Lime + 0.065305^*Organic Matter + 0.0809364^*CEC - 0.273767^*Clay - 0.321222^*Ombro + 0.15906^*Eto + 0.214134^*Giacobe - 0.23289^*\alpha + \beta - pinene - 0.269601^*sabinene + 0.328806^*myrcene - 0.0464061^*\delta - 3-carene - 0.196524^*limonene - 0.29596^*\beta - phellandrene - 0.308632^*terpinen-4-ol + 0.140899^*\beta - caryophyllene + 0.333913^*germacrene-D.$

| 1. α+β-Pinene | |
|-------------------------------------|---|
| 2. Sabinene | |
| 3. Myrcene | |
| 4. δ-3–Carene | a. Electrical Conductivity Extract Saturation |
| 5. Limonene | b. Active lime |
| 6. β-Phellandrene | c. Organic matter |
| 7. Terpinen-4-ol | d. Cation Exchange Capacity |
| β-Caryophyllene | e. Clay |
| 9. β-Germacrene-D | f. Exchangeable Sodium Percent |
| A. Ombrothermic Index | |
| B. ETo (Evapotranspiration P-M) | |
| C. IG (Giacobbe index) | |

5. Conclusions

The five sampling locations presented well defined bioclimatic and edaphic characteristics that could be related with specific EO profiles. These results suggest that the EO composition of *P. lentiscus* L. depends on bioclimatic and edaphic factors rather than on the geographical origin of the samples. Even more, the edaphic factor, seems to have more influence than the bioclimatic factor on EO composition.

Some general trends have been established based on the obtained results: the Calcaric Arenosol (Saler) with a high evapotranspiration index could be associated with a high sesquiterpenic fraction, (germacrene-D and β -caryophyllene, mainly). For the Luvisol (Borrell and L'UII) high amounts of α + β -pinene were found to be related with sub-humid bioclimatic conditions and clayey and fertile soils (high CEC and OM levels). This last requirement was also observed in relation to the myrcene content, but with drier climatic conditions and

calcium carbonate-dominated soil process (Haplic Calcisol) (high level of carbonates and active lime) (Lliria). The rest of the hydrocarbon monoterpene and oxygenated monoterpene (terpinen-4-ol, mainly) fractions could be related to drier climates and soils which are not calcareous or very fertile such as the Eutric Regosol (Segart).

Finally, given the most suitable composition related to specific and useful biological activities, new research of controlled environmental factors (soil features and bioclimatic conditions) should be conducted in order to define the best conditions to manage industrial crops of *P. lentiscus* L.

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