Università
degli Studi
di Ferrara

INTERNATIONAL DOCTORAL COURSE IN
“EARTH AND MARINE SCIENCES (EMAS)”
CYCLE XXXV

Coordinator: Prof. Paolo Ciavola

Innovative geomaterials for a sustainable agriculture and Agri-Food traceability

Scientific Disciplinary Sector (SDS) GEO/09

Years 2019/2022

Candidate

Dott. Valeria Medoro

Valeria Medoro

Supervisors

Prof. Massimo Coltorti

Prof. Carmela Vaccaro

Co-supervisor

Dott. Giacomo Ferretti

Contents

List of abbreviations:

AN: Ancona site

ANOVA: ANalysis Of Variance

BA: Bioaccumulation Index

BG: Brisighella site (Ravenna)

BN: Bertinoro site (Forlì-Cesena)

BO: Bologna site

CWSI: Crop Water Stress Index

VWC: Volumetric Water Content

E: Evapotranspiration

GS: Stomatic Conductance

Pn: Net Photosynthesis

CA: Kaolin

CEC: Cation Exchange Capacity

CNT: control treatment

DM: Dimethoate; application of 100% chemical pesticide

EA-IRMS: Elemental Analysis-Isotope Ratio Mass Spectrometry

FA: Fatty Acid

FAME: Fatty Acids Methyl Esters

FUE: Fertilizer Use Efficiency

GC-IRMS: Gas Chromatography-Isotope Ratio Mass Spectrometry

IAEA: International Atomic Energy Agency

IC: Ion Chromatography

ICP-MS: Inductively Coupled Plasma Mass Spectrometry

MN: Montiano site (Forlì-Cesena)

NS: no water stress

NUE: Nitrogen Use Efficiency

PCA: Principal Component Analysis

PDO: Protected Designation of Origin

PE: Pescara site

PGI: Protected Geographical Indication

PLS-DA: Partial Least Square-Discriminant Analysis

RE: Recovery Efficiency

REE: Rare Earth Elements (in this work I also include Rb, Sr, Zr, Nb, Hf, Ta, Th and U)

S: water stress

SF: Spyntor Fly; it is the traditional foliar treatment with Spynosad and Spyntor fly

SL: San Lazzaro di Savena site (Bologna)

TC: Transposition Coefficient

TSG: Traditional Specialties Guaranteed

VIP: Variable In the Projection

ZA: Enriched Zeolitite; it is a zeolitite enriched with NH_4^+

ZEO: zeolite treatment; it is the treatment with 50% less of N-fertilizers and the application of zeolitite

ZN: Natural Zeolite

ZN-DM: it is the treatment with 50% of dimethoate and natural zeolite

Abstract

In the last decades, the overuse and low efficiency of fertilizers and chemical products led to an increased interest in innovative and eco-friendly strategies for improving the sustainability of agricultural practices. The use of geologic materials, such as zeolite-rich tuffs (also known as zeolitites), is now a well-established technique able to face this daunting challenge. Thanks to their properties, zeolitites are considered soil improvers and have been found to improve the efficiency of a large spectrum of products, from synthetic and organic fertilizers to also pesticides. Zeolitites are often applied in the soil to retain and slowly release nutrients and water, improving plant growth and reducing nutrient losses in the environment. They are also sprayed as particle film on plant leaves and fruit to protect them from pests and environmental stresses. Although their common application as soil amendment in many crops, zeolitites have not yet been tested in olive growing and grapevine where large quantities of N fertilizer and pesticides are still used.

However, strategies which imply the modification of the soil physico-chemical properties, or which foresee the application of particle films on plant organs may also induce significant changes in the geochemical fingerprint of the plant. For this reason, in this thesis, the geochemical and isotopic compositions were studied in plants and final products with a food traceability approach to identify the geochemical footprint in relation to the cultivar and soil and thus to discriminate the fertilization management and univocally identify the product according to the agricultural practices. The geographical origin of Agri-Food products was so investigated due to the key role of the pedology, geologic and climatic features of the experimental sites.

The research developed in the following PhD thesis falls within the priorities of Cluster 6 “Food, Bioeconomy, Natural Resources, Agriculture and Environment” (Horizon 2023-2024) whose focus is the “restoring Europe’s ecosystems and biodiversity, and managing sustainably natural resources to ensure food security and a clean and healthy environment”.

The work has been organized into two main experiments focusing on olive growing and grapevine. Many aspects were considered, such as the importance and influence of agricultural practices and the traceability of the final products.

EXPERIMENT A ON OLIVE GROWING: To improve the sustainability and productivity of modern agriculture, it is mandatory to enhance the efficiency of Nitrogen (N) fertilizers with low-impact and natural strategies, without impairing crop yield and plant health. To achieve these goals, the ZeOliva project experimented the use of a zeolite-rich tuff as a soil amendment to improve the efficiency of the N fertilizers and allow a reduction of their inputs. The results of three years of experimentation performed in three different fields in the Emilia-Romagna region (Italy) are presented. In each field, young olive trees grown on zeolite-amended soil (<50% of N-input) were compared to trees grown on unamended soil (100% N-input). Soils and leaves were collected three

times every year in each area and analyzed to monitor the efficiency of the zeolite treatment compared to the control. Vegetative measurements were performed along with analysis of pH, Soil Organic Matter and soluble anions in soil samples, whereas total C and N, C discrimination factor and N isotopic signature were investigated for both soils and leaves. Besides some fluctuations of nitrogen species due to the sampling time (Pre-Fertilization, Post-Fertilization and Harvest), the Total Nitrogen of leaves did not highlight any difference between treatments, which suggests that plant N uptake was not affected by lower N input in the zeolite treatment. Results, including vegetative measurements, showed no significant differences between the two treatments in all the observed variables, although the control received twice the N-input from fertilization. Based on these results, it is proposed that zeolite minerals increased the N retention time in the soil, allowing better exploitation by plants which led to the same N uptake of the control notwithstanding the reduction in the N inputs. The use of zeolite-rich tuff in olive growing thus allows a reduction in the amount of fertilizer by up to 50% and improves the N use efficiency with many environmental and economic benefits.

Moreover, food quality and safety have become of great interest, with a consequent demand for geographical identification of agri-food products and eco-friendly agricultural practices. For this reason, geochemical analyses of soils, leaves and olives from two areas in the Emilia-Romagna Region (Italy), Montiano (MN) and San Lazzaro (SL) were performed aiming at identifying geochemical fingerprints able to determine 1) univocally the locality of provenance and 2) the effect of different foliar treatments (control which means no foliar treatments, organophosphate insecticide as the dimethoate, and alternating of natural zeolitite and dimethoate in MN; Spinosad+Spyntor fly, natural zeolitite and NH_4^+-enriched zeolitite in SL). Principal Component Analysis (PCA) and Partial Least-Squares Discriminant Analysis (PLS-DA, including index values of the Variable Importance in Projection -VIP analysis-) were used to discriminate between localities and different treatments. Bioaccumulation and Translocation Coefficients (BA and TC) were studied to evaluate differences in the uptake of trace elements by plants. The PCA performed on soil data highlighted a total variance of 88.81%, allowing a good distinction between the two test sites. Leaves and olives PCA showed that using trace elements it is possible to discriminate different foliar treatments (total variance: 95.64% and 91.08% in Montiano; 71.31% and 85.33% in San Lazzaro of leaves and olives, respectively), and an excellent result is also obtained in the identification of their geographical origin (87.46% of leaves and 83.50% of total variance of olives). PLS-DA of all samples gave the largest contribution to the discrimination of different treatments and geographical identification. Among all elements, only Lu and Hf were able to correlate soil, leaf, and olive for geographical identification through VIP analyses, but also Rb and Sr were significant in the plant uptake (BA and TC). For the discrimination of different foliar treatments, Sm and Dy were identified in MN site, whereas Rb, Zr, La and Th correlated leaves and olives from SL. Based on trace element analyses, it can be put forward that 1) the geographical origin could be discriminated and 2) different foliar treatments applied for crop protection can be recognized, which means, reversing the reasoning that each farmer can develop a method to pinpoint his product through geochemical markers.

Finally, olive oils were analyzed to identify the geographical fingerprint by the carbon signature of Fatty Acids (FAs). Olive oils from six sites in the north and centre of Italy (BO-Bologna, BG-Brisighella, SL-San Lazzaro di Savena, MN-Montiano, PE-Tocco da Casauria and AN-Jesi) were analyzed by GC-IRMS. The effects of different foliar treatments were tested on the isotopic composition of FAs from some sites (Kaolin, Natural Zeolitite and Control in BO; Spinosad+Spyntor fly, Natural Zeolitite and NH_4^+-Enriched Zeolitite in SL; Dimethoate, Natural Zeolitite and Control in MN). PCA in olive oil data explained 69.61% of the total variance for all areas and PLS-DA showed the best separation of BG and SL from the other sites, while MN highlighted less differentiation compared to all other experimental fields. Among the most abundant FAs in olive oils, stearic acid C18:0 was the best discriminator in VIP analysis, followed by linolenic acid C18:3w6,9c and oleic acid C18:1w9c. The results of olive oils PCA to verify the influence of foliar treatments on the FAs composition showed a total variance of 89.35% in SL, 71.98% in MN and 71.54% in BO, proving that agricultural practices can influence the isotopic fingerprints of olive oil FAs. Zeolitite as foliar treatment probably caused a direct or indirect effect on olive oil, probably providing a direct input of different C signature and/or improving the plant photosynthesis and consequently altering the $\delta^{13}C$ ratio.

EXPERIMENT B ON GRAPEVINE: The combination of agricultural practices and water stress conditions were also investigated in grapevine to discriminate their effects on the final products. In particular, the use of zeolitite as soil amendment (ZEO) was tested both on normally irrigated plants and on those under water stress (S). The results obtained on 2-year-old grapevines grown under a shade canopy environment in Susegana (CREA, TV, in the north of Italy) were presented for the 2020 agronomic year. Soils and leaves were collected three times and analyzed to verify if some differences occurred thanks to the use of zeolitite. Soil basic parameters and Total Carbon (TC), $\Delta^{13}C$ and geochemical analysis on trace elements were measured on soil and leaf samples. Bioaccumulation Coefficient (BA), Crop Water Stress Index (CWSI), Volumetric Water Content (VWC) and gas exchanges were determined. Statistical analyses, such as PCA and PLS-DA including VIP analysis, were performed to distinguish between groups. Water stress conditions highlighted their influence on plant physiological status (CWSI, VWC and gas exchange) and TC in leaves, as well $\Delta^{13}C$, with lower values in water-stressed than well-watered plants. Zeolitite showed meaningful effects mainly in the geochemical composition of both soils and leaves. Zeolitite probably introduced some elements in soil, for example, Sr was twice in zeolitite treatments compared to other treatments, while in leaf samples only Rb was meaningfully higher thanks to the use of zeolitite in soil, also verified by BA coefficient. PCA and PLS-DA of all samples confirmed the separation due to the agricultural practices (89.29% for soils and 88.81% for leaves) and highlighted the importance of zeolitite in the geochemical composition much more than the plants

water conditions. VIP analysis pointed out Rb, Sr and Ta as the discriminators of the agricultural practices in both soil and leaf samples. Based on these results, it is suggested that the use of zeolitite in grapevine allows discrimination in the trace elements composition of leaf samples, which could be detected in grapes, but further investigation is needed to verify this hypothesis.

1. Introduction

1.1 Requirement of new sustainable agriculture practices

The low Fertilizer Use Efficiency (FUE) is one of the main causes of the altered equilibrium of agro-ecosystems (Mustafa et al., 2021) and it is responsible for relevant economic losses for farmers (Basso et al., 2016; Drechsel et al., 2015). The role of N-based fertilizers is to provide an adequate amount of N to the plants and grant a good yield. However, after the addition of fertilizers to the soil, N is generally not efficiently uptaken by the plant, but it is lost in the surrounding environment through several pathways, causing the degradation of the soil, water compartments and increasing the concentrations of Greenhouse Gas (GHG) in the atmosphere (Dawson et al., 2008; Peng et al., 2006). As pointed out by Drechsel et al. (2015) and Chien et al. (2016), the apparent recovery efficiency (RE) of N by crops is lower than 55%. For this reason, to guarantee a crop yield able to sustain the future demands in terms of food for the population, there is an urgent need to: (1) improve the efficiency of agricultural practices, (2) reduce the N losses in the environment as harmful greenhouse gasses or leachates, and (3) reduce the use of N based fertilizers without compromising the productivity of the plants (Cataldo et al., 2021; Ferretti et al., 2017b; Sharma and Bali, 2018; Soares et al., 2012). Moreover, reducing the amount of fertilizers, especially those produced by synthetic processes such as urea (Shi et al., 2020), represents a great saving in terms of energy and exploitation of non-renewable resources. Improving FUE would also have great value for organic farming, which is known to have a limited set of products with low-N content available for fertilization purposes. The reduction of chemical fertilizers and pesticides is one of the biggest issues on which the EU council (Green Deal plan) is working. New strategies are being studied to decrease, by 2030, the amount of soil for crops, increase the biodiversity, grow up organic farming by +25% and preserve soil, water and human health. Bremmer et al. (2021) reported that if the Green Deal objectives are not reached, the future scenario will be characterized by lower production, price increases, fewer European exports and more imports of agricultural products from outside Europe. Cluster 6 from Horizon programme 2023-2024 “Food, Bioeconomy, Natural Resources, Agriculture and Environment” included the targets and objectives of the Green Deal core, in particular, the farm-to-fork and biodiversity strategies, zero pollution efforts and climate action. One of the specific topics of the Horizon programme is the development of sustainable, productive, environment-friendly and resilient farming systems providing consumers with affordable, safe, traceable, healthy and sustainable food while minimising pollution and pressure on ecosystems and emissions of greenhouse gases.

Thereby, the development of eco-friendly practices to reduce the use of fertilizers while improving their efficiency is necessary to increase production in terms of quality and quantity and to guarantee human and environmental health, accordingly to the UE directions (Water Framework Directive 2000/60/CE, Directive 2009/128/CE for the pesticide use and Nitrates Directive 91/676/EEC). To reduce the leaching losses and increase the efficient use of the N-fertilizers, the N retention in the soil represents the key to limiting the amount of N lost in the environment by giving "more time" to the plants to exploit the N storage in the soil (N reservoir).

Furthermore, in olive growing, the defence of olive plants from pests, fungi, and insects (i.e. sooty mould, Verticillium albo-atrium, olive moth, cochineal, and especially the Bactrocera oleae Gmelin or olive fly) that cause the early loss of olives and some negative effects in oil composition, is one of the most important problems. In Italy, the Bactrocera oleae Gmelin or olive fly is the worst pest and it is spread in all Mediterranean areas, causing an huge loss in production and the subsequent increase in the use of chemical pesticides to fight it. On the other hand, the use of pesticides as foliar treatment causes environmental and health problems and their efficiency is often linked to climate variables. Moreover, the growth in the use of pesticides and fertilizers is not sustainable due to their long degradation time and their toxic residues in the final products (olive and oil) which compromise its quality and safety. The olive fly is even more critical for organic farms that have scarce tools to control it. For these reasons, EU encourages the introduction of innovative techniques and integrated defence to reduce the use of pesticides (Directive 2009/128/CE). The interest in developing natural and chemical-free alternatives to pesticides, such as organic agrochemicals or the use of geologic material, is rising: European Commission Regulation EC No. 432/2012 established a list of permitted health claims made on foods. Therefore, to guarantee human and environmental health, natural zeolitites were used as a foliar treatment to reduce the chemical pesticides to fight the olive fly.

From a geological point of view, zeolitite (or zeolitic-tuff) means a tuff rock (Kesraoui-Ouki et al., 1994) with more than 50% of zeolite minerals. Zeolite mineral has a 3D structure with a framework consisting of $[SiO_4]^{4-}$ and $[AlO_4]^{5-}$ tetrahedra. This mineral has an extra framework formed by weakly-bounded exchangeable cations and H_2O molecules that occupy their open cavities (channels and cages). Zeolite minerals can be described by their common formula (1):

$$M_x M'_y N_z \left[T_m T'_n \ldots O2_{(m+n)-\varepsilon}(OH)_{2z}\right](OH)_{br}(aq)_p * qQ \qquad (1)$$

Where $\varepsilon = 0,1,2,3...$; M and M' are exchangeable and non-exchangeable cations, respectively; N represents non-metallic cations; (aq) are chemically bonded water; Q is sorbate molecules; T and T' are Si^{4+} and Al^{3+} (Reháková et al., 2004).

The Si^{4+} can be substituted by Al^{3+} ions, which produces a negative charge framework balanced by alkali and alkaline earth cations. These cations have relative free mobility through the extra-framework and they can also be replaced by other ions available in the surrounding environment ("Cation Exchange" substitution process).

The Si/Al ratio is one of the main parameters that define the Cation Exchange Capacity (CEC) of zeolite minerals. On the whole, the most common CEC (meq g^{-1}) are 3.0-3.5 for phillipsite and chabazite, 2.0-2.3 for clinoptilolite and 1.8-2.0 for mordenite.

Thanks to their structure and chemistry, zeolites have the following main properties: i) low density and large extra-framework linked spaces, ii) high cation exchange capacity (CEC), iii) reversible dehydration, iv) high crystallinity, v) molecular sieve and vi) catalytic properties (Coombs, 2001; Moshoeshoe et al., 2017).

Zeolite minerals formation is generally linked to high alkalinity (pH>7), low temperature (<300°C) and low-pressure conditions. They can occur in some igneous or metamorphic rocks as primary minerals thanks to the crystallization at high temperatures in their fractures by hydrothermal alteration or by metamorphism processes; near the surface, at low temperatures, they can crystallize under specific chemical gradients of the interstitial waters. Zeolites from hydrothermal alteration are only 5-10% of the bulk rocks so they only have a scientific value. However, in most cases, zeolites are secondary phases and are associated with volcanic rocks (pyroclastic sediments) under diagenesis (Delkash et al., 2015; Gottardi, E. & Galli, 1985). These pyroclastic sediments have often a significant glass content that influences the chemistry of zeolites: i) a zeolite with a high Si/Al ratio is formed from a SiO_2 rich glass whereas ii) a zeolite with a low Si/Al ratio from "basaltic" glass. The chabazite-zeolite applied in this experiment is included in the low Si/Al group and thereby it is characterized by a very high CEC. Zeolites from diagenesis of pyroclastic sediments may occur with a high content of bulk rock and for this reason, they also have an economic and industrial relevance (the rock with more than 50% zeolite minerals can be called zeolitite or zeolitic-tuff).

Although there are many natural zeolites (IZA Commission on Natural Zeolites (iza-online.org), last updated on 24th of June 2022) which differ in structure and properties, the most common and studied are clinoptilolite, mordenite, chabazite, phillipsite and erionite (Galli and Passaglia, 2011).

These highly reactive minerals have unique properties that make them very useful for many purposes, including agriculture (Ferretti et al., 2020; Reháková et al., 2004; Rotondi et al., 2021). When used as a soil amendment, zeolitites are useful to improve the capacity of the soil to retain nutrients and water, improving plant growth (Colombani et al., 2016, 2015; Di Giuseppe et al., 2016; Malferrari et al., 2013; Passaglia, 2008; Perez-Caballero et al., 2008). With this method, plants can uptake nitrogen more efficiently and the nitrogen losses in the surrounding environment can be significantly reduced (Coltorti et al., 2012; Eslami et al., 2018; Faccini et al., 2018; Ferretti et al., 2018, 2017a). In this context, their use as an inorganic amendment is becoming popular in many crops, such as maize, apple trees, sorghum, corn and soy to cite some examples (Campisi et al., 2016; Faccini et al., 2018; Ferretti et al., 2019, 2017a; Milosevic and Milosevic, 2009). Many works have been carried out about Nitrogen management in olive growing and its effects on the plants' growth (Fernández-Escobar, 2011; Fernández-Escobar et al., 2012, 2004; Ferreira et al., 2020; Othman and Leskovar, 2019). Nevertheless, zeolitite application in olive growing has been a subject of investigation in a few works (Lopes et al., 2020; Perez-Caballero et al., 2008), even though excessive dosing of mineral fertilizers are often used. Fernández-Escobar (2011) reported that up to 200 kg-N/ha can be applied to adult olive trees, although this quantity can satisfy their N demand for years.

This work aims at testing the use of zeolitite in olive-growing as a soil amendment for granting lower inputs of N-based fertilizers. It is expected that the zeolite minerals may influence the N dynamics in the soil, promoting a prolonged permanence of this nutrient and reducing the losses in the surrounding environment. This should be reflected in a more efficient uptake by plants and therefore in the possibility to significantly reducing the N inputs while maintaining crop quality and yield.

In this framework, the results of three years of experimentation in three different experimental sites are presented. An Italian chabazite-rich zeolitite quarried in Sorano was used as a soil amendment in olive growing to reduce the fertilizer N input by 50% compared to common practices. During the experimentation, vegetative measurements were performed, and samples of soil and leaves were collected three times every year to measure a series of chemical parameters (including soil basic parameters, inorganic anions, nitrogen speciation and N-C stable isotopes), to account for differences between treatments and to evaluate the efficiency of this practice.

1.2 Need for Agri-Food traceability and safety

The identification of a chemical and isotopic fingerprint of agrifood products to identify their origin has been a matter of interest for the scientific community for years (Llorent-Martínez et al., 2011; Pepi et al., 2018). Traceability allows tracing food, feed or substance that will be transformed into food, following all stages of production, processing, and distribution, according to Regulation EC N°178/2002 (European Parliament, 2002). Traceability defines the properties of that product and the characteristics of the environment from which it comes, as a kind of identity card for the food product (Regattieri et al., 2007). It is indeed well known that the elemental composition of some agri-food products, such as wine (Baxter et al., 1997; Jakubowski et al., 1999; Thiel et al., 2004) as well as coffee, tea, olive oil, and fruit juice (Ogrinc et al., 2003) is deeply correlated to the geochemical composition of the soil where they have been cultivated (Gouvinhas et al., 2016; Mansour-Gueddes et al., 2022; Nasr et al., 2021; Tyler, 2004), to the agricultural practices (Bontempo et al., 2009; Pepi et al., 2016a) and to the local environmental conditions affecting the biogeochemical cycling.

To determine the geochemical profiles of Agri-Food products, the most common analytical method is Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The geographically and geologically sensitive parameters, such as trace elements, have significant relevance to characterize and subsequently identify the origin of a given food product. Therefore, these methods are frequently used to identify the geographical origin as reported by several authors (De la Guardia and Garrigues, 2015; Mir-Marqués et al., 2016; Pepi et al., 2019, 2018, 2017; Rodrigues et al., 2011). Most of all, Rare Earth Elements (REE) have recently been suggested as potential traceability markers for Agri-Food products (Aceto et al., 2019; Punturo et al., 2018; Turra, 2017), due to their considerable interest in industry, agronomy and food science (Turra, 2017; Tyler, 2004). Rare Earth Elements (REE) are a group of 17 trace metals of the periodic table which includes the lanthanide group and yttrium (Dukov, 2007). Despite their name, REEs represent the fifteenth most abundant component of the Earth's crust and they cannot be considered rare (Hu et al., 2006). They have similar chemical and physical properties (Christopher H. Evans, 1990), but the ionic radius of Sc is much smaller than other REEs and for this reason, it is not classified as LREE and HREE. Generally, REEs have trivalent oxidation states, except Ce (4+) and Eu (2+) in some environments (Wang and Liang, 2015). Despite their similarity, due to their differences REEs are generally divided into two groups: Light REE (LREE) from La to Nd and Heavy REE (HREE) from Ho to Lu, including Y (Dukov, 2007; Long et al., 2012; Tyler, 2004). Sometimes, also a third group can be added between LREE and HREE and it is called Middle REE (MREE). MREE include Sm, Eu and Gd. LREE are generally more soluble and less able to form complexes than HREE (Cantrell and Byrne, 1987; Goldschmidt, 1937; Henderson, 1984).

In the last years, the demand for REE in the market of green technology is increased, especially in industry, medicine and agriculture (Eriksson and Olsson, 2011). REEs are part of several daily devices, such as lighters, television sets, computers, medical technology, nuclear engineering, automobile industry, military devices, spacecraft (Kato et al., 2011) and wind turbines. In fact, a growing number of emerging alternative energy technologies (e.g. electric and hybrid vehicles, car catalysts, energy-efficient lighting, wind power) and digital equipment (e.g. flat panel displays, disk drives, digital cameras) contain REE-bearing components (Haxel GB, Hedrick JB, 2002; Long et al., 2012). REEs are also used in medicine, such as Gd-bearing for the contrast of media for magnetic resonance imaging or La carbonates for the treatment of renal disease. With wide use and few suppliers, REEs are in the list of critical raw materials in the European Union (European Commission 2010) and in the United States (Norman et al., 2014). Raw material is labeled 'critical' when there is a risk for supply shortage and the impacts of REE in the economy are higher compared with most of the other raw materials (European Commission 2010). The rapid growth of REE has led to an exponential increase of global REE and their use leads to environmental contamination. REEs are therefore considered as an emerging pollutant (Kulaksiz and Bau, 2011; Yang et al., 2009). In general, REEs have shown low concentrations in soils, plants, water and atmosphere, but they maybe accumulated in such environments due to anthropogenic inputs as well as enter in the food chain (Turra, 2018). In China, Korea, Japan, Australia, Switzerland, the Philippines (Wang et al., 2008) and in others countries a mixture of REE in fertilizers has been used in agriculture to improve the nutrition of plants (Hu et al., 2004). Agricultural supplies may have REE in their composition, mainly in fertilizers which are rich in phosphate (Turra et al., 2011), and some plants may accumulate these elements (De França et al., 2011; Ozaki and Enomoto, 2001; Robinson et al., 1958, 1938; Turra et al., 2013). Also other industrial activities can release substances into the soil, air, water and plants. REE can persist in the environment, causing bioaccumulation in biota, water pollution and chronic toxicity (Chua, 1998; Laveuf et al., 2012). REE may enter the human body mainly by the ingestion of contaminated food and inhalation of dust carried by air (Chua, 1998). There are many studies that have shown REE toxicity effects in animals like rats (Christopher H. Evans, 1990), crayfish (Eyerusalem Goitom, 2011), shrimps (Wang et al., 2000), guinea pigs and in human beings. Given that REE can accumulate in soils because of their overuse in industry and agriculture (Haxel GB, Hedrick JB, 2002; Hu et al., 2004; Kovaříková et al., 2019; Tyler, 2004; Zhou et al., 2017), it is necessary to know the REE concentration in soil due to the different pedogenic processes, including the physico-chemical characteristics and anthropogenic changes of soil. Therefore, it is important to investigate the influence of soil on the absorption and fractionation of REE by plants and their translocation to fruits (Tyler, 2004). In the last decade, some studies were carried out about the absorption of chemical elements from the soil to the plant and their bioaccumulation (Amorós et al., 2013; Buscaroli, 2017; Concas et al., 2015; Pepi et al., 2016b; Punturo et al., 2018; Vystavna et al., 2015). Other studies investigated the toxicity of heavy metals and other environmental contaminants, especially in grapes and wines (Alagić et al., 2015; Bravo et al., 2017; Protano and Rossi, 2014), and the capacity of plants to uptake elements from soil and roots, to evaluate the transport of chemical elements into the food chain (Buscaroli, 2017; Pošćić et al., 2020). Some of these contaminants and heavy metals can be carried into the food chain and affect the quality of Agri-Food products.

For these reasons, the EU is aiming to guarantee eco-friendly and human-healthy agricultural practices. Most of all, Regulation (EC) N° 178/2002 assures the general principles and requirements of food law, establishing the procedures in matters of food safety and the basis for a high level of protection of human health in relation to food. It is applied to all stages of production, processing and distribution of food and feed. Moreover, the "European White Paper on Food Safety" (12 January 2000) aims to achieve the highest level of health protection for the consumers of European food, and also IAEA (2011) plays a key role in assuring food safety. In addition to EU regulations, many countries have established their own traceability regulations. The Italian Standards Institute (UNI) has enacted specific legislative measures. To cite an example, the UNI 11020:2002 defines principles and requirements for the development of traceability systems in agro-industries. Other legislative acts have been introduced in several European countries, such as France, Spain and Greece, but they primarily relate to quality issues rather than food safety.

The EC regulations N. 509/2006, 510/2006, 1898/2006, defend consumers in respect of the protection of geographical indications and designations of origin for agricultural products and foodstuffs, through controls that define the safeguard of the Protected Designation of Origin (PDO), the Protected Geographical Indication (PGI) and the Traditional Specialties Guaranteed (TSG). The more representative food products with PDO are wines, cheeses, olive oils, honey, beers, meats and potatoes (Sacco et al., 2009) because there is a long tradition associating these products with specific areas (Luykx and van Ruth, 2008). Italian oils are considered among the best in the world thanks to regional microclimates and autochthonous cultivars. They need a supply chain traced by geochemical markers to guarantee high-quality products, especially organic oils (i.e. DOP and IGP products labelled) and obtained with sustainable agronomical practices. In this framework, the evidence of provenance has become the key to solving the problem, accordingly to national legislation and international standards. Consequently, the determination and amount of trace and REE elements concentration (Benincasa et al., 2008, 2007; Brescia et al., 2005) of a particular and unequivocal region can be the key to providing the unique compositional fingerprint for characterizing the geographical origin of olives, and, in a near future, of olive oils (Kelly et al., 2005).

In the last decade, there was an increasing interest in organic products and environmental-friendly methods of production (Luykx and van Ruth, 2008). As a result of this new global trend, the impact evaluation of the new agricultural practices on the geochemical composition of final products has become mandatory. At the same time, the high quality of food products depends on the geographical origin (Drivelos and Georgiou, 2012) and their safety and geochemical fingerprints strongly depend on the management of agricultural practices. To guarantee food safety, environmental protection and a crop yield able to sustain the future demands in terms of food for the population, there is an urgent need to improve the efficiency of agricultural practices and/or decrease the amount of fertilizers needed to reach a certain level of production (Ferretti et al., 2017b; Soares et al., 2012). For this reason, various strategies are being adopted to reduce the use of synthetic products and within them, the use of geologic materials represents a natural and eco-friendly way to face this issue. In this context, in the last decades, volcanic tuffs rich in zeolite minerals (zeolitite, if the zeolite content is above 50%) have been tested as soil amendment for many crops, such as maize, apple trees, sorghum, corn and soy to cite some examples (Campisi et al., 2016; El-Sherpiny et al., 2020; Faccini et al., 2018; Ferretti et al., 2019, 2017a; Milosevic and Milosevic, 2009; Minardi et al., 2020; Rodrigues et al., 2021). Although their common use as a soil amendment to reduce the overuse of fertilizers, zeolite minerals have never been exploited as a foliar treatment. In the case of olive growing, zeolitites as foliar treatments can contrast the olive fruit fly by creating a natural highly reactive barrier on the leaves and the fruits (De Smedt et al., 2015). In the olive growing, the defence of olive plants from pests, fungi, and insects (i.e. sooty mould, *Verticillium alboatrum*, olive moth, *cochineal*, and especially the *Bactrocera oleae Gmelin* or olive fly) that cause the early loss of olives and some negative effects in oil composition, is one of the most important problems. In Italy, the olive fly is the worst pest because it is spread in all Mediterranean areas, causing the subsequent increase in the use of chemical pesticides to fight it. On the other hand, the use of pesticides as foliar treatment causes environmental and health problems and their efficiency is often linked to climate variables. At the same time, olive oil consumers have become more demanding in terms of hedonistic product quality and food safety because they associate the product with a healthy lifestyle. As a consequence, the olive oil producers had to provide oil with high qualitative features and reduce chemicals with high environmental and health damage. The growth in the use of pesticides and fertilizers is not environmentally friendly due to their long degradation time and their toxic residues in the final products (olive and oil). For these reasons, UE encourages the introduction of innovative techniques and integrated defence to reduce the use of pesticides (Directive 2009/128/CE). The reduction in the farmer's dependency on pesticides has a key role in the "Food, Bioeconomy, Natural Resources, Agriculture and Environment" (Horizon 2023-2024), with the aim of sustainable and climate-friendly farming systems, providing economic, environmental, and social and health benefits.

To guarantee human and environmental health, natural zeolitites were used as a foliar treatment to reduce the chemical pesticides to fight the olive fly (ZeOliva project financed by the Ministry of Agricultural, Food and Forest Policies).

This study aims at univocally linking the final products (olives) to their soils and treatments, through the geochemical fingerprints (trace elements and REE) that allow identifying their provenance and use of biological practices in open-field experiment. The influence of environmental features, as well as soil types, was investigated to discriminate the area of origin, testing the traceability tools in a more limited area, where the distinction between two areas nearby is much more difficult than distinguishing products from areas far away. The influence of different agricultural practices was also investigated for each site: in each experimental field, the use of zeolite-rich tuff was compared with other traditional foliar treatments commonly used in olive growing. The influence of these new agricultural practices on the geochemical composition was studied on leaves and olives. Moreover, research on the Fatty Acids composition in oils was conducted.

Generally, Fatty Acids (FAs) rarely occur as free molecules in nature but are usually found as components of many complex lipid molecules such as fats (energy-storage compounds) and phospholipids (the primary lipid components of cellular membranes). FAs consist of a chain of an even number of C-atoms, with H-atoms along the length of the chain and a carboxyl group (-COOH) at the other end. The carboxyl group makes it an acid (carboxylic acid). If the carbon-to-carbon bonds are all single, the acid is saturated; if any of the bonds is double or triple, the acid is unsaturated and is more reactive. The most frequent FAs are the 16- and 18-carbon fatty acids (C16:0 and C18:0, respectively), also known as palmitic acid and stearic acid. Both palmitic and stearic acids occur in the lipids of the majority of organisms. In animals C16:0 is more than 30% of body fat, whereas is between 5 and 50% in vegetable fats, abounding especially in palm oil. On the other hand, some vegetable oils (e.g. cocoa butter and shea butter) are rich in C18:0.

Fatty acids have a huge range of commercial applications. They are used not only in food products, but also in soaps, detergents, and cosmetics. Some skin-care products contain fatty acids, which can help preserve healthy skin appearance and function. FAs, most of all omega-3 fatty acids, are also commonly sold as dietary supplements because many animals, including humans, cannot synthesize linoleic acid (an omega-6 fatty acid) and alpha-linolenic acid (an omega-3 fatty acid). Those fatty acids are essential for cellular processes and the production of other necessary omega-3 and omega-6 fatty acids (which derive from linoleic and alpha-linoleic acids). Omega-6 and omega-3 fatty acids are formed in the body from their parent fatty acids but not always at levels needed to maintain optimal health or development. For all these reasons, they are called essential fatty acids and they must be taken in through the diet.

Olive oils are rich in both saturated and unsaturated FAs (Christy and Egeberg, 2006; Wabaidur et al., 2016), but the common FAs are tridecanoic acid (C13:0), myristic acid (C14:0), pentadecanoic acid (C15:0), palmitic acid (C16:0), margaric acid (C17:0), stearic acid (C18:0) (saturated acids), oleic acid (C18:1) and linoleic acid (C18:2) (unsaturated acids) (*American Oil Chemists' Society, AOCS Official Method*, 2006a, 'American Oil Chemists' Society, AOCS Official Method', 2006b). But the composition of the FAs varies in the oils and they contain a high proportion of unsaturated FAs compared to saturated FAs (Christy and Egeberg, 2006). The proportion of oleic to linoleic acids varies in edible oils. For example, olive oil contains 85% oleic acid compared to 39% in soya oil. At the same time, soya oil contains 54% linoleic acid compared to 9% in olive oil (Christy and Egeberg, 2006). FAs were well studied to i) determine quality indices of the oils during the production, storage and trading (Prentki and Madiraju, 2012; Soto-Vaca et al., 2013), ii) optimize the oil refining and authenticity, iii) control their degradation under some circumstances (Díaz and Borges, 2012), iv) detect the adulterations of high quality with cheap oils (Christy and Egeberg, 2006) and v) correlate FAs to the quality and authenticity of the oils. For this reason, a study of FAs in olive oils became necessary to univocally identify the isotopic signature of C atoms in FAs chains.

On the other hand, grapevine represents a huge crop in the Made in Italy production and Agri-Food traceability. Nitrogen (N) is one of the most important nutrients in the grapevine and its deficit can limit the growth and development of the plant (Bell and Henschke, 2005). It also controls the vigour, yield and berry quality (Habran et al., 2016; Jackson et al., 1993), as well as the ratio of vegetative growth, which results in an alteration in berry growth and a change in the chemical composition of the berry, the must and the wine (Echtebarne et al., 2010; Kliewer and Dokoozlian, 2005). For these reasons, N is the most commonly nutrient applied in doses between 40 and 80 kg/ha per year (Janick, 1992). N can be assimilated in various parts of the plant, such as the roots, trunk, stem, leaves and berries (Wermelinger et al., 1991) in different forms, like nitrate, ammonium, urea, glutamine and arginine. In the last years, N uptake mechanisms have been well-studied but only a few works investigated how grapevine accumulation is influenced by the different N forms and whether berry quality is influenced by the N form (Lang et al., 2018).

Additionally, in the wine sector, Italy is the second-largest producer in the world after France and exports a third of its production. The wine supply chain is considered extremely elaborated because of the several steps required to make the final product. For this reason, in the wine chain, the first step for traceability is to trace the primary raw material, the grapes, moving from the vine-growing to the vinification and the further steps of the supply chain (bottling and distribution). The traceability of grapes is of great interest to producers, distributors and consumers because the geographical origin of these products and their environmental features are the discriminant aspects in the production of a competitive product on the market. Furthermore, Italian wines need to differ in quality on the international market: Federvini reported that 60% of Italian export wine is for Europe, 31% for America and 7.75% for Asia. To avoid competition due to the price, there were concluded trade agreements for the defence of Made in Italy (such as Protected Geographical Indication PGI, and Protected Designation of Origin DOP labels). Over the years, it is increased the demand for high-quality wines (Geographical Indication, IG), which has been evidenced by the willingness to spend on these products: large-scale retail trade (GDO) registered an increase of +5% in the first eight months of 2019. In Italy, nowadays there are more than 526 wines GI labelled and consumers have focused on the wine/terroir relationship. This attention to the geographical origin of the products led to food and wine tourism, proving the consumers' desire to combine the products and the experience linked to the discovery of the territory. The chemical characterization of grapes is a requisite to identify univocally the geographical origin of this product. The concentration of chemical elements plays a fundamental role in the development of a new approach for “chemical traceability”, due to the close link between the environmental features (geological factors, mineralogy and geochemistry of vineyard soils, physico-chemical properties of soils, bioavailable concentration, weathering processes…) and their influences in the plant uptake of nutrient and non-essential elements (Protano and Rossi, 2014). Major elements and REE are used in several studies as fingerprints of wine origin (Almeida and Vasconcelos, 2003; Fabani et al., 2009; Galgano et al., 2008; Kment et al., 2005; Rodrigues et al., 2011; S. et al., 2001; Taylor et al., 2003). Many studies have investigated the geographical origin of grapes using chemical composition (Censi et al., 2014; Cugnetto et al., 2014; Galgano et al., 2008; Mercurio et al., 2014; Protano and Rossi, 2014; Vystavna et al., 2015), or have evaluated translocation and accumulation of chemical elements in the soil-plant system (Abreu et al., 2012; Angelova et al., 1999; Leita et al., 1998). But only few works are performed about the behaviour of trace elements in the soil according to irrigation levels, the related Index of Bioaccumulation (BA) and the relationship between trace elements and soil management (Pepi et al., 2016a). The purpose of this experiment was to evaluate the effects of different soil management practices and their combination on major and trace elements concentrations in soil, describing the index of bioaccumulation (BA) related to these elements in leaves and grape berries. Furthermore, geology, climate, geography and anthropic factors, as well as their interaction with different grape varieties, may cause the transfer of trace elements from soil to plant (Flexas et al., 2001; Vystavna et al., 2015). Moreover, the grape chemistry, as well as the winemaking techniques, influence the elemental profile and quality of the wine. For all these reasons, this method has largely been used as a chemical tool to identify the geographical origin of wines.

This work aims at studying grapes of *Cv* Glera under different agricultural practices (such as water stress conditions and soil amendment) to verify how treatments could influence the geochemistry and the physiological status of grapevines. The effects of these practices were studied on soils and leaves.

2. Goals

On one side, this work aimed at testing innovative geomaterials to evaluate the effects of these new agricultural practices to reduce the N-fertilizers and the environmental impact caused by their overuse. To achieve this goal, three years of experimentation (in three different experimental sites) in which an Italian chabazite-rich zeolitite was used as a soil amendment in olive growing to reduce the fertilizer N input.

On the other side, this work aimed at tracing the final products (olives and olive oils) from their soils and leaves, to determine the geochemical and isotopic fingerprints due to their origin from different geographical areas and the use of different agricultural practices (the same chabazite-rich zeolitite was applied in micronized form as foliar treatment in adult olives to fight the olive fly). Moreover, different agricultural practices (water use, application of zeolitite, N-enriched fertilization) and their effects were tested on grapes in order to trace these products depending on the agricultural practices.

For these reasons, the work has been organized into two experiments:

1. Experiment A on olive growing: olive trees test. In the first part of this experiment, agricultural practices and their effects on the olive plants were tested, which means the study of zeolitite as a soil amendment and foliar treatment. Therefore, a statistical approach was performed to trace olives from their soils according to their agricultural practices and their geographical origin. Finally, an olive oil characterization was performed for the traceability of the final product.
2. Experiment B on grapevine: grape test. In the first year of the experiment, a different combination of the use of zeolitite in soil and water application was tested to trace the geochemical composition in grapes according to agricultural practices.

3. Material and Methods

3.1 Zeolitite employed in the experiment

For all the experiments, a chabazite zeolite was chosen due to its high CEC which makes it very useful for agricultural and industrial applications (Mumpton, 1999). Chabazite has a framework structure consisting of a stacked sequence of 6-rings in the order AABBCC..., forming double 6-rings at each apex of the rhombic unit cell (Figure 1). The largest channels perpendicular to [001] (hexagonal setting) are confined by eight-membered rings (aperture 3.8 x 3.8 Å). The framework topology of the CHA structure type is rhombohedral, R3m. Generally, Mazzi and Galli (1983) suggested that chabazite may have randomly arranged domains with perfect (Si, Al) ordering.

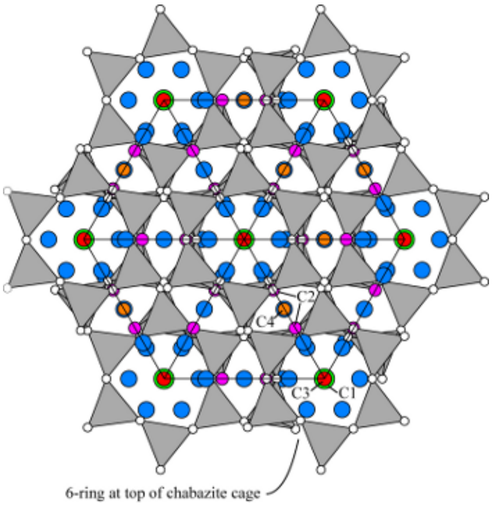

Figure 1. Crystal structure of chabazite zeolite. Four channel cation sites were distinguished: C1 (green in the figure) outside the double 6-ring and bonded with 3 oxygen atoms of the ring, C2 (magenta) near the 8-ring port of the chabazite cage and bonded to three framework oxygen atoms, C3 (red) near the center of the cage, not bonded to the framework, and C4 (orange) at the center of the 8 ring. C1 and C3 generally contain most of the extra-framework cations (Mazzi and Galli, 1983). Blue circles represent positions of H_2O molecules. (*www.iza-online.org/natural/Datasheets/Chabazite/chabazite*)

The chabazite zeolitite (NZ) used in this experimentation is a volcanic tuff quarried in Sorano (42°41’20.65” N; 11°44’26.29” E, Grosseto, Italy), provided by Verdi S.p.a. company. This specific zeolitite has been widely studied in open-field and laboratory tests (Campisi et al., 2016; Faccini et al., 2018; Ferretti et al., 2020, 2018, 2017a; Galamini et al., 2020; Malferrari et al., 2013; Mastrocicco et al., 2016; Medoro et al., 2022). The NZ was composed of nearly 70% of zeolite minerals, mainly K-rich chabazite, which gives this NZ a very high CEC (Table 1). The main characteristics of the NZ are reported in Table 1. Quantitative Phase Analysis of zeolitite (QPA) was estimated by X-Ray Diffractometry (XRD) X’Pert PRO MPD (PANalytical, Netherlands) at the University of Padova.

The zeolitite was back-loaded to minimize crystallite-preferred orientations and ZnO at 20% (w:w) was added to the sample for the quantification of the amorphous phase (volcanic glass). The fundamental parameters approach, accordingly to the Profex-BGMN implementation (v.5.0.1), was used in the Rietveld refinements and the associated QPA (Cheary et al., 2004; Doebelin and Kleeberg, 2015) (Cheary et al., 2004; Doebelin and Kleeberg, 2015), while the BGMN database was used to identify the structure of crystals.

Table 1. Main properties of the chabazite-rich zeolitic tuffs. PSD, particle size distribution; QPA, quantitative phase analysis; TZC, total zeolitic content; CEC, cation exchange capacity. Tot.CEC refer to the total CEC. The CEC data were obtained by XRD analysis at University of Padova and by the selling company for the CHA zeolitic tuffs, respectively.

chabazite-zeolitic tuff

PSD µm	*(%)*	**QPA**	*(%)*	**CEC** *(mEq/g)*	
500-250	0.0	Chabazite	71.3	Ca^{2+}	1.46
250-125	0.0	K-Feldspar	13.8	K^{+}	0.60
125-63	0.13	Phillipsite	4.7	Na^{+}	0.07
63-3.9	57.1	Phlogopite	2.4	Mg^{2+}	0.04
< 3.9	42.7	Quartz	1.2		
		Glass	6.7		
		TZC	76.0	Tot.CEC	2.17

The total CEC of the chabazite zeolite is 2.17 meq g^{-1} and it is enriched in exchange potassium (K^{+}) and calcium ions (Ca^{2+}), followed by Na^{+} and Mg^{2+} in a small amount.

The NZ was employed in a granular form, with a particle size ranging between 3 and 6 mm, as soil amendment. A micronized NZ with average particle size between 6-10 µm was used as foliar treatment.

3.2 Experiment A: olive tree test

The experiment is part of the project ZeOliva, financed by the Italian Ministry of Agricultural, Food, and Forestry Policies (MIPAAF) and led by the University of Ferrara (Unife) and the National Research Council of Bologna (CNR). It aims at reducing the use of fertilizers in olive growing by exploiting the properties of natural zeolite minerals as a soil amendment and at fighting the olive fly by using natural zeolite as foliar treatment. This work led to a publication in Land Journal (MDPI) available to the link Land | Free Full-Text | Reducing Nitrogen Fertilization in Olive Growing by the Use of Natural Chabazite-Zeolitite as Soil Improver (mdpi.com).

This work will focus on the use of zeolitite to 1) improve the soil properties through the zeolitite as soil amendment, in order to investigate the efficiency of new sustainable agriculture practices and 2) discriminate Agri-Food products for traceability purposes thanks to the application of zeolitite as foliar treatment. The researches followed different experimental set-up, as described in the next chapters.

3.2.1 Zeolitite as soil amendment in agricultural practices and experimental set-up considerations

To evaluate the effects of the NZ in increasing the efficiency of fertilizers and allowing a reduction in fertilizer input, two treatments were compared in 5-year old olive trees:

1) CNT: 100% fertilizer N input and unamended soil (common practice);
2) ZEO: 50% fertilizer N input and addition of natural zeolitite as soil amendment (500 g added to each plant at planting phase in 2016–2017 at a depth of 30–40 cm).

The fertilization reduction was performed according to the fertilization plan adopted at each field by the owner company. Different fertilizers were used in each field as well as slightly different amounts (see detailed description for each site). The experimentation started in February 2019. The monitoring lasted three years and was replicated in three different experimental fields located in various provinces of the Emilia-Romagna region, suited to olive growing (Figure 2).

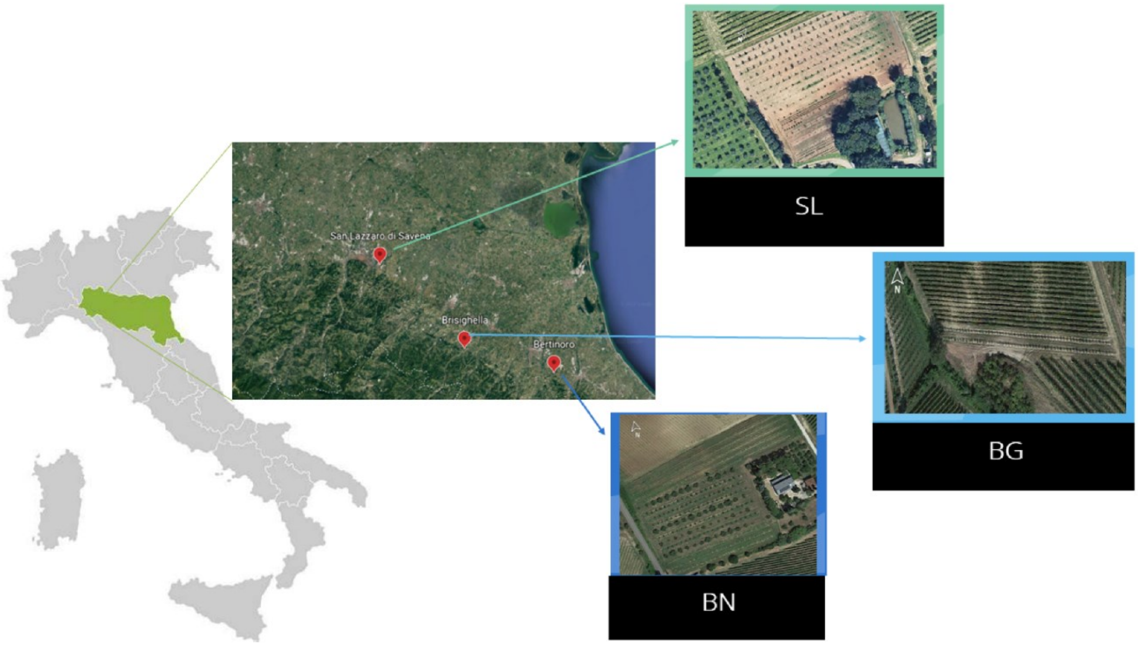

Figure 2. Geographical location of San Lazzaro (SL), Brisighella (BG) and Bertinoro (BN) experimental fields in Emilia-Romagna region (Italy).

At each site, three olive trees were selected randomly per each treatment (ZEO and CNT) to serve as replicates. Soil and leaf samples were collected three times each year (2019, 2020 and 2021): 1st before the fertilization (Pre-Fert) during the vegetative rest, 2nd after the fertilization (Post-Fert) during the vegetative recovery and 3rd at the olive harvest (Harvest) at each site (Figure 3). Soil samples were collected from the 0–30 cm soil layer and about 10 cm from the plant stem with an Eijkelkamp (Ø 30 mm × 500 mm) auger. Three subsamples were collected for each tree and mixed to form a single representative composite sample. For each tree, more than 20 leaves were randomly collected at each sampling. The total number of samples processed every year was 108 (considering 2 treatments, 3 experimental sites, 3 time points, 3 replicatres, resulting in 54 soil and 54 leaf samples); over the 3 years, a total of 324 samples was processed.

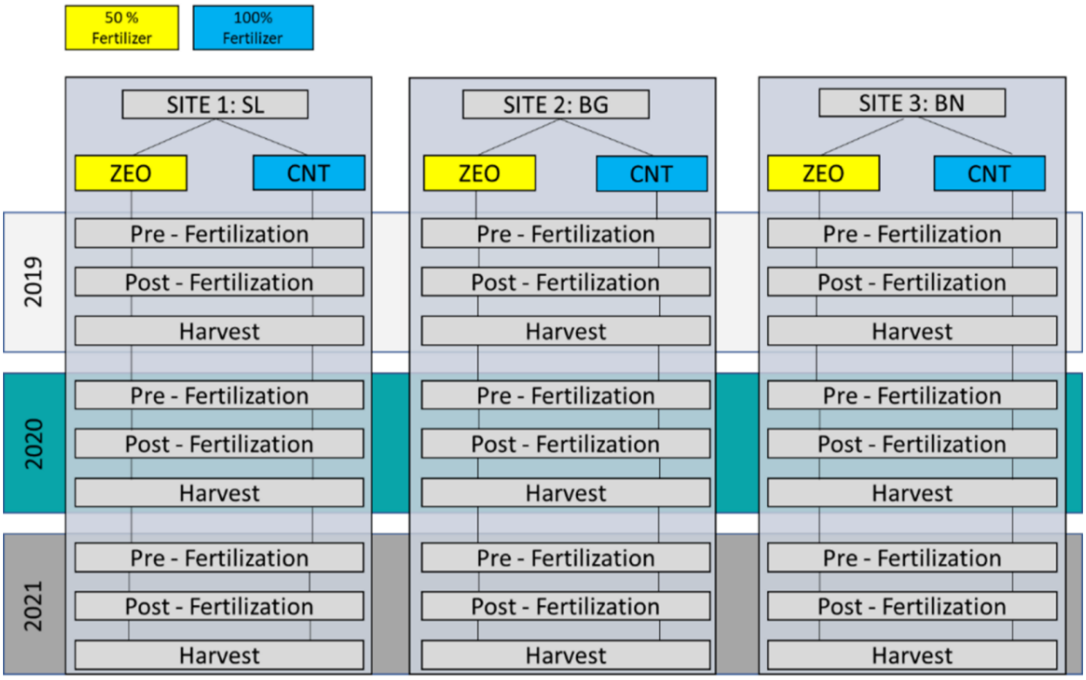

Figure 3. Experimental set-up of three fields: San Lazzaro (SL), Brisighella (BG) and Bertinoro (BN). At each site ZEO and CNT treatments were tested; three replicates were sampled three times per year (Pre-Fertilization, Post-Fertilization and Harvest).

Site 1: San Lazzaro di Savena (SL)

The "SL" experimental field is located within the Bologna province and belongs to the "Azienda Agricola Bonazza" (organic regime). According to the soil map of the Emilia Romagna Region (GeoViewer—Geoportale), the soil belongs to the unit CDV1 that is mainly represented by Hypocalcic Vertic Calcisol soils according to the World Reference Base for Soil Resources (2022)

(‘GeoViewer — Geoportale’, n.d.; Tarocco, A. and Aprea, 2021). The orchard consists of 6 rows of olive trees: 3 rows of Cv Montecapra, Montebudello and Farneto whose soil was treated with zeolitite at transplanting (ZEO) and 3 rows of the same Cv whose soil was left untreated (CNT). Then, 500 g of NZ were added to the ZEO treatment at transplanting (in March 2017) assuring contact with plant roots. Since 2019, organic fertilization has been halved (50% of fertilizer/year) only in the zeolite thesis (ZEO), whereas 100% of fertilizer was applied in the CNT.

In 2019, the fertilization was completed with an NP organic fertilizer (Phoenix NP, N 6%, C 2%) followed by a manure application in June for a global input of approximately 40 kg N/ha which corresponds to 118 g of N per tree in the CNT. Half of these dosages were used in the ZEO treatment.

In May 2020, Biouniversal fertilizer (N 11% and C 40%) was applied at a dosage of 37 kg N/ha in the CNT, corresponding to 55 g of N per tree, whereas half of the dosage was applied in the ZEO treatment.

In March 2021, Agriazoto11 (N 11% and C 39%) was applied in the same quantity as 2020. The olive grove was rainfed. The mean temperature for the overall period (2019-2021) was 15.3 °C and precipitation was approximately 635 mm, with the maximum rainfall recorded in 2019 (866 mm) and 570 mm and 468 mm for 2020 and 2021, respectively (Pavan, 2022, 2021; Pavan and Marletto, 2020). Three plants per treatment were randomly selected for soil and leaf sampling.

Site 2: Brisighella (BG)

The “BG” experimental field is located within the Ravenna province and belongs to “Azienda Agricola Giorgia”. BG soil belongs to the cartographic unit BAN3/SOG according to the Emilia Romagna Region soil map that is mainly represented by Haplic Regosols (World Reference Base for Soil Resources (2022)) (‘GeoViewer — Geoportale’, n.d.; Tarocco, A. and Aprea, 2021). The olive grove consists of two olive rows of Cv Nostrana di Brisighella and three plants of both CNT and three ZEO treatments were selected for the sampling of soils and leaves. As in the SL area, 500 g of zeolitite per olive tree were added to the soil of the northern row (in May 2016) to create the ZEO treatment. Since the transplant, chemical fertilization has been halved (50% of fertilizer/year) in the ZEO treatment, whereas CNT received 100% of fertilizer.

In March 2019, the fertilization was performed using an organic-mineral fertilizer (Cosmo N 13%) using 100 kg N/ha in the CNT which corresponds to 185 g of N per tree and half of the dosage in the ZEO treatment. In June 2020, 50 kg N/ha of NH4NO3 (N, 34%) were applied to the CNT corresponding to 93 g of N per tree while half of the dosage was applied in the ZEO treatment. In March 2021, 37 kg N/ha of Urea (N, 46%) per tree were used in the CNT (corresponding to 69 g of N per tree) while half of the dosage was applied in the ZEO treatment. The orchard was irrigated with no differences between CNT and ZEO. The BG site showed the highest precipitation in 2019 (1072 mm), whereas during 2020 and 2021, precipitations were between 600 and 650 mm. The average temperature was 13.7 °C (2019–2021) but in July 2020 and August 2021, peaks of 40 °C were reached, surpassing the average temperature for that period in the last decade (Pavan, 2022, 2021; Pavan and Marletto, 2020).

Site 3: Bertinoro (BN)

The "BN" experimental field is located within the Forlì-Cesena province and belongs to the "Azienda Agricola Tenute Unite". The soil belongs to the cartographic unit DEM/BAN3/DOG0 that is mainly represented by Haplic Cambisol according to the World Reference Base for Soil Resources (2022) ('GeoViewer — Geoportale', n.d.; Tarocco, A. and Aprea, 2021). The orchard is made up of different olive cultivars among which Colombina, Correggiolo Pennita and Capolga di Romagna were chosen to conduct the experiment. The set-up was similar to SL and BG sites: three plants were selected for CNT and three for ZEO treatments for soil and leaf sampling; in November 2016 the soil was amended with 500 g of zeolitite (ZEO treatment).

The BN site was managed with a considerably lower N input with respect to the other 2 sites. In 2019, Dermazoto (N 11% o, C 80%) was applied in March. The second fertilization was completed in June 2019 under the same conditions for a total of 7.5 kg N/ha (corresponding to 11 g per tree) while half of the dosage was used in the ZEO treatment.

The same fertilizer was applied also in May 2020 and March 2021, respectively at dosages of 5.6 and 6.7 kg N/ha, corresponding to 16.5 g and 19.8 g of N per tree, whereas half of the dosage was used in the ZEO treatment. The orchard was rainfed. BN recorded a mean temperature of 14.6 °C, aligned with the average temperature of the previous years. The mean precipitation for 2019–2021 was 595 mm, with the highest values recorded in 2019 (823 mm) and slightly lower than 500 mm in 2020 and 2021 (Pavan, 2022, 2021; Pavan and Marletto, 2020).

3.2.2 Zeolitite as foliar treatment in agricultural practices and experimental set-up considerations

This work led to a submitted manuscript nowadays under review in a scientific journal. The experiment was carried out in two different experimental fields located in the Emilia-Romagna region (Italy), suited to olive growing. The fields belong to two different companies and are located in two different areas: Montiano (MN, Forlì-Cesena, 44°5'2,04"N, 12°18'19,44"E) and San Lazzaro di Savena (SL, Bologna, 44°28'22"N, 11°24'10"E).

MN soil was characterized by a predominant clay-silt fraction, while SL soil was characterized by an important sand fraction and was classified as sandy-loam/clay-loam (Martelli et al., 2009). Montiano originated from "Argille Azzurre" Formation (FAA), which includes grey-blue mudstone and silty clay, often with stratifications due to the bioturbation; furthermore, their internal structure can be very complicated due to the great lithological variability linked to the sedimentary basin and the local coarse contribution (Martelli et al., 2009). On the other hand, San Lazzaro site is mainly composed of “Formazione delle Sabbie Gialle di Imola” (IMO), which means a fine and very fine sandy formation, with less coarse sands and unusual and discontinuous mudstone intercalations. The youngest deposit in this area is a member of “Sintema Emiliano-Romagnolo Superiore” (AES) of the Quaternary age: the typical “Subsintema di Bazzano” (AES6) in San Lazzaro includes gravel-sand from alluvial terrace (Martelli et al., 2009). According to the soil map of the Emilia-Romagna Region ((https://geo.regione.emilia-romagna.it/cartografia_sgss/user/viewer.jsp accessed on 22nd August 2022) MN soil belongs to the unit AGE1/CEL3 which correspond to Haplic Cambisols and Hypocalcic Leptic Calcisols, while the SL soil belongs to the unit CDV1 that is mainly represented by Hypocalcic Vertic Calcisol soils according to the World Reference Base for Soil Resources (2022) (‘GeoViewer — Geoportale’, n.d.; Tarocco, A. and Aprea, 2021).

At each site, soil, leaf and olive samples were collected during the olive harvest that occurred in October 2019. The year 2019 was characterized by extreme thermal fluctuation and variability of precipitation: dryness, heavy rainfalls and high temperature characterized the first months, frequent precipitations and low temperatures distinguished the spring and increasing temperatures were recorded in the summer months. The report of IdroMeteoClima Emilia-Romagna revealed that temperatures rose around 15°C in one week between May and June and maximum precipitation was reached in May and November while the lowest precipitations were recorded in June. Moreover, some extreme events occurred over the year. In SL the average annual temperature was 15.4°C and the precipitation 866mm, while MN had a mean of 14.3°C and 795mm of precipitation (Pavan and Marletto, 2020).

At each site, three adult olive trees cv Correggiolo were selected randomly per treatment (see the following description) to serve as replicates. Both sites provide DOP Colline di Romagna olive oil production. Both orchards were rainfed.

The MN orchard (belonging to Azienda Agricola Egisto Bocchini) was cultivated under integrated farming. The orchard consists of 5 plants per row and 8 total rows, which means 40 olive trees. The following foliar treatments, accordingly to the company treatment plans, were tested:

i) 100% dimethoate (ROGOR 400 SP/Epik SL), the traditional foliar treatment applied by the company (DM);
ii) alternation of natural zeolitite and dimethoate, with a reduction of 50% of the dimethoate application (ZN-DM);
iii) negative control, without any foliar treatment (CNT).

The application of DM and ZN-DM was made when the active infestation threshold exceeded 3% from July to the olive harvest. DM was applied by dissolving 100 mL of dimethoate in 100 L of water. In the ZN-DM treatment, 800 g of micronized zeolitite was added to 100 L of water; zeolitite and dimethoate were applied separately every 15/20 days (7 total treatments).

The SL experimental field belongs to the “Azienda Agricola Bonazza”. The orchard consists of 6 rows of olive trees with 5 plants per row (30 olive trees). The SL orchard was cultivated under organic farming and three different foliar treatments were compared, according to the company management plan:

i) natural micronized zeolitite (ZN);
ii) NH_4^+-enriched zeolitite (ZA);
iii) Spinosad+Spyntor fly on traps (SF) representing the common practice in organic regime.

Seven foliar treatments were applied from July to October (at olive harvest) every 15/20 days (7 total treatments). In ZN treatment, 800 g of micronized zeolitite were added to 100 L of water. In ZA treatment, 400 g of natural micronized zeolitite were mixed with 1 L of NH_4^+ solution (3000 ppm of NH_4^+) to produce the NH_4^+-enriched zeolitite. 400 g of NH_4^+-enriched zeolitite and 800 g of natural zeolitite were added to 100 L of water. In SF treatment, traps with 1-1.2 L of Spinosad+Spyntor fly were dissolved in 4 L of water/ha and applied at 50% of the plants, as suggested by the producers.

The geographically and geologically sensitive parameters, such as trace elements, have significant relevance in order to characterize and subsequently identify the origin of a given food product. Therefore, these methods, expecially Rare Earth Elements, are frequently used to identify the geographical origin as reported by De la Guardia & Garrigues, 2015; Marqués et al., 2016; Pepi et al., 2017, 2018, 2019; Rodrigues et al., 2011. Recently, REEs have been suggested as potential traceability markers in Agri-Food products (Aceto et al., 2019; Punturo et al., 2018; Turra, 2018), due to their considerable interest in industry, agronomy and food science (Turra, 2018; Tyler, 2004). REEs can accumulate in soils because of their overuse in industry and agriculture (Haxel GB, Hedrick JB, 2002; Hu et al., 2004; Kovaříková et al., 2019; Tyler, 2004; Zhou et al., 2017), so it is necessary to know the REEs concentration in soil due to the different pedogenic processes, including the physico-chemical characteristics and anthropogenic changes of soil. Therefore, it is important to investigate the influence of soil in the absorption and fractionation of REEs by plants and their translocation to fruits (Tyler, 2004).

For these reasons, there was studied trace elements, especially REEs, in order to test if any difference occurred between samples from different areas and/or with different treatments. The analyses were performed from soils to olives, using a statistical approach for the fruit traceability by trace elements. All samples, both soils, leaves and olives refer to the 2019 agronomic year to avoid variability due to the different climate conditions among agronomic years.

3.2.3 Olive oil characterization: a statistical approach for the final product traceability

The research was led in collaboration with the Boku University of Tulln an der Donau (Vien, Austria) during the period abroad for the PhD. The experiments were carried out in the laboratories of the

Department of Forest and Soil Sciences of Tulln and thanks to the collaboration with the “Stable Isotope Group” of the Institute of Soil Research (IBF).

To characterize the olive oil composition, the fatty acids profile or fatty acids composition (saturated, unsaturated and monounsaturated) was analyzed qualitatively and quantitatively by gas chromatography.

In this work the “omega” notation was applied to indicate FAs:

$$\mathrm{C}x\colon y\omega c$$

Where C*x* is the number of C-atoms in the FA, *y* is the position of double bounds, ω represents the unsaturated FAs and *c* or *cis* means that fatty acids have two hydrogen atoms attached to the double bond on the same side of the carbon chain.

Gas chromatography (GC) and GC with mass spectrometry (GC-MS) have been used for the indirect analysis of FAs with improved resolution in the last decades (Ackman, 2002; Brondz, 2002; Jalali-Heravi and Vosough, 2004).

In order to discriminate and classify the olive oils from different geographical areas, a study of the isotopic composition of FAs was performed. The C isotopic signature (δ^{13}C) from the fatty acids chain was analyzed by Gas Chromatography coupled with an Isotopic Ratio Mass Spectrometry (GC-IRMS).

Olive oils from different regions of Italy were chosen for the experiment (Figure 4). For some of them, also δ^{13}C signature was studied as a result of different treatments in olive growing (SL and MN treatments are already described in the previous chapter “Zeolitite as foliar treatment”).

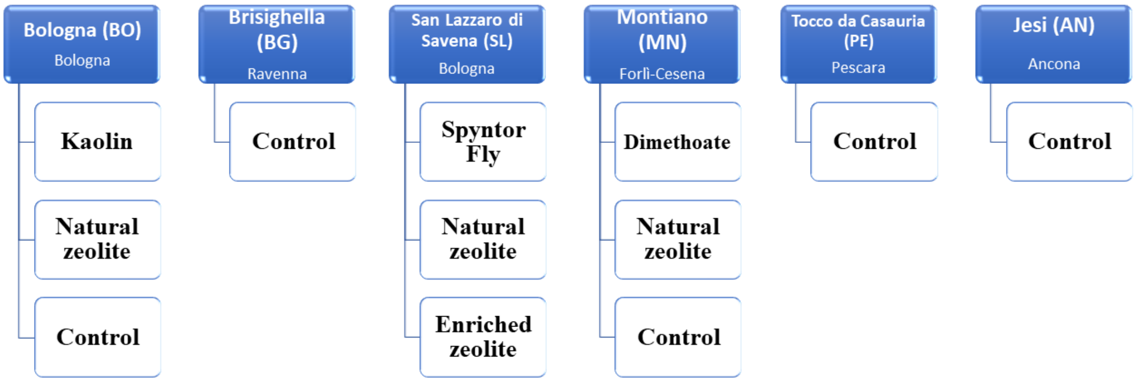

Figure 4. Experimental set-up of six fields: Bologna (BO, 44°29′38″N, 11°20′34″E), Brisighella (BG, 44°13'21"36 N, 11°46'32"88 E), San Lazzaro di Savena (SL, 44°28′17.64″N, 11°24′17.49″E), Montiano (MN,

42°38′47″N, 11°13′25″E), Tocco da Causaria (PE, 42°12'54"N, 13°54'53"E) and Jesi (AN, 43°37′N, 13°31′E). At some sites different treatments were tested as foliar treatment, whereas in other experimental fields only control (no treatments) was studied; data referred to the 2019 year.

All samples refer to the 2019 agronomic year to avoid variability due to the different climate conditions among agronomic years, as already described in the previous chapter for olive traceability.

3.3 Experiment B: grape test

The experiment was led in collaboration with the Council for Agricultural Research and Economics (CREA) of Conegliano (45°53′14″N, 12°17′49″E, Treviso, Veneto region, Italy). All the experiments were carried out in a shade canopy environment (a system of various structures and coverings that can reflect, block and control the amount of sunlight hitting the plants) in Susegana (45°51'25"56 N, 12°15'16"20 E Treviso, Veneto region, Italy), in which different treatments were tested on young grapes of *Vitis Vinifera Cv Glera* (2-years old).

3.3.1 Zeolitite and water use effects on vineyard

The experiment was set up in the agronomic years 2019-2020 when the 2-years old vineyard was too young to be in production. For this reason, all the analyses were done in soils and leaves, but not in grapes.

The combination of zeolitite as soil amendment and water stress conditions in plants was investigated in order to evaluate their contribution to the discrimination of the final product based on the treatment. In particular, there were tested i) the use of zeolitite as soil amendment (ZEO) and ii) the effects of water stress (S); these combined practices have led to the following treatments (Figure 5):

1. Zeolite in soil and water stress in plants (Szeo);
2. Zeolite in soil without water stress, which means normal irrigation conditions (NSzeo)
3. Unaltered soil and plant water stress (S)
4. Unaltered soil without water stress, which means normal irrigation conditions (NS)
5. Control (CNT), which means without any treatments.

Twelve replicates were collected for each treatment, except for CNT which had four replicates.

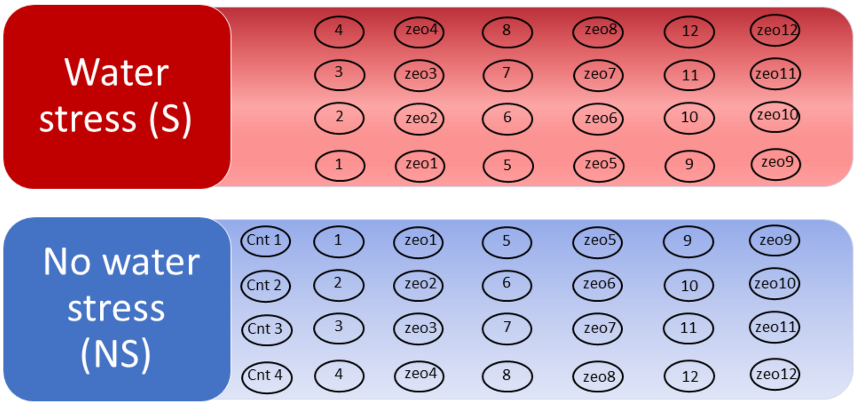

Figure 5. Experimental set-up of vineyard in shade canopy environment (52 pots). In the upper part, all plants were under water stress (24 pots); in the lower part, all plants received normal irrigation (no water stress, 24 pots). For each irrigation condition, 12 pots were amended with zeolitite (Zeo) and 12 without (unaltered soil). All 48 pots received the fertilization with digestate. 4 pots were for the control treatment (CNT) and have not received any fertilization. Numbering indicates replicates of the same treatment.

Preliminary analyses were done to estimate the amount of N-fertilizer (digestate) for each pot by Kjeldhal method and EA-IRMS.

In July, 2 L of sandy soil from the alluvial plain of Susegana were mixed with 50 mL of digestate in all 48 plots (except 4 plots of CNT treatment). In 24 plots of them also zeolitite (already described in detail in the previous chapter) was added as soil amendment in the 10% w/w. Three samplings were planned during the year to evaluate the adsorption of N during the time: at fertilization, after two weeks from fertilization and at harvest (in September).

A complete investigation of soils and leaves were planned. Basic soil parameters were measured, such as pH and Soil Organic Matter (SOM); soils were also analized by IC for the measurement of principal anions; soil and leaf samples were analized by ICP-MS for the determination of major, minor and in trace elements and high metals; also EA-IRMS was applied for all samples to determine the δ^{15}N and δ^{13}C signatures and the study of elementar N and C.

3.4 Analytical techniques

3.4.1 Soils and leaves

Textural analysis

Particle size analyses of four samples per area were done to characterize the soil texture of each area: samples were manually divided into quarters and opposite quarters were chosen for the analyses. To remove the organic matter, soils were treated with H_2O_2 (16 times the volume of the sample); then they were left for 24 hours at environmental temperature. The sandy fraction was separated from the silty-loam fraction by a 63 μm sieving: the coarse one was weight after being dried at 105°C for 24 hours; the finest parts (silt and loam) were left to sediment in water in a plastic decanter of 5 L and then quantified with an X-ray sedigraph (Micromeritics 5100) at standard conditions, a dimensional range from 0.0884 mm to 0.00049 mm, and a standard density value of 2.7 g/cm3. 0.5 L of Sodium Esamexaphosphate with a low concentration (0.5%) was added to the finest fractions to simplify the grain scatter. All data obtained from the textural analyses were used for the USDA classification by Sedimcol software.

Chemical analyses

Both for olive tree and grape experiments, soil samples were collected at 30 cm of depth and about 10 cm from the plant stem with an Eijkelkamp (Ø 30mm x 500mm) auger. They were air-dried and sieved at 5 mm before further analysis. 20 leaves were sampled randomly and collected from each plant. Leaf samples were dried at 60°C for 72 hours and grounded with an electric grinder until obtaining fine powder.

Soil samples were extracted with H_2O Milli-Q (high purity) at 1:10 ratio (weight/volume), to measure soluble anions and pH. After shaking for 1 h at 150 rpm in closed plastic tubes, the supernatant was separated by centrifugation at 4000 rpm for 4 minutes and filtered with 0.45 μm Cellulose Acetate Abluo syringe filters (GVS Filter Technology). pH was measured with a pH electrode connected to an automatic titrator unit 877 Titrino-Plus (Methrom, Italy). Soil H_2O extracts were analyzed by Ionic Chromatography (IC) with an ICS-1000 Dionex equipped with AS9-HC 4x250 mm anion column, AG9-HC 4x50 mm guard column, ADRS600 suppressor and AS-40 autosampler for the determination of soluble anions (F^-, Cl^-, NO^{2-}, Br^-, NO^{3-}, PO_4^{3-}, SO_4^{2-}). Calibration was performed with certified Thermo Fisher Scientific standards. Moreover, Soil Organic Matter (SOM) was measured: 0.5 g of soil was heated at 550°C to weigh the mass losses of the samples.

The Total Nitrogen and Carbon (respectively TN and TC) and the respective isotopic signature ($\delta^{15}N$ and $\delta^{13}C$) of soil and leaf samples were acquired with a Vario Micro Cube Elemental Analyser (EA) (Elementar, Langenselbold, Germany) connected to an Isoprime 100 mass spectrometer (IRMS) (Isoprime, Cheadle, UK) operating in a continuous-flow mode. The EA-IRMS was calibrated with synthetic Sulfanilamide (provided by Isoprime Ltd) and Carrara Marble (cross-calibrated at the Institute of Geoscience and Georesources of the National Council of Researches of Pisa) standards.

In order to study the traceability of Agri-Food products from their soils and leaves, samples were prepared for ICP-MS analysis. soil samples were air-dried and grounded before further processing. Accordingly to Mastrocicco et al., 2016, 0.2 g of sample were subjected to acid digestion by adding 6 mL of HF (40%, Suprapur®, Merck KGaA, Darmstadt, Germany) and 3 mL of HNO_3 (65% in distilled water, Suprapur®, Merck KGaA, Darmstadt, Germany) in a 50 mL-Teflon digestion vessel, 43 × 60 mm (VWR International, Milan, Italy). Then, samples were heated at 190°C until complete drying. This procedure was repeated 3 times with different volumes of acids: 3 mL of HF and 3 mL of HNO_3 (2nd cycle), 4 mL of HNO3 (3rd cycle) and 2 mL of HNO_3 (4th cycle). The final solution was diluted to 100 mL with highly purified MilliQ® water. Soil samples were analyzed by ICP-MS to determine trace elements and REE using a Thermo Electron Corporation X series spectrometer (Thermo Fisher Scientific).

0.4 g of leaf sample were subjected to acid digestion by the addition of HNO_3 (65% in distilled water, Suprapur®, Merck KGaA, Darmstadt, Germany) and H_2O_2 (37% in distilled water, Suprapur®, Merck). In a 50-mL Teflon digestion vessel, 43 × 60 mm (VWR International, Milan, Italy) 4 mL of HNO_3 and 3 mL of H_2O_2 were added to the solution and heated at 150°C until complete drying. The procedure was repeated by adding 3 mL of HNO_3 and 2 mL of H_2O_2 and heating at the same temperature until complete drying. Finally, the dried residue was resuspended in 3 mL of HNO_3 diluted with highly purified MilliQ® water before being analyzed by ICP-MS.

3.4.2 Digestate

The digestate wastewater was sampled in a biogas production plant located near Ferrara city (San Biagio village, Italy 44°58'47″ N; 11°85'86″). It resulted from the anaerobic digestion of dairy manure and triticale. Suspended solids as vegetable residues and the heavier granular fractions were separated by centrifugation (4000 rpm for 10 minutes) and discarded. The collected liquid part was analyzed by EA-IRMS to measure the $\delta^{15}N$ and Kjeldhal system for the NH_4^+-N and Total Kjeldahl Nitrogen (TKN).

For the determination of digestate isotopic signature, 1 L of digestate was mechanically mixed with 60 g of granular chabazite zeolitite (~6% w/w) for 2 hours. The resulting zeolitite was heated at 40-50°C for one night and powdered in agate mortar. The same analyses was also performed with unaltered digestate on a ashless paper filter Whatman-110 mm diameter (Cytiva, United States). 10 mg of enriched-zeolitite and 8 mg of unaltered digestate were analyzed by EA-IRMS.

Digestate samples were extracted with H_2O Milli-Q (high purity) at 1:50 ratio (weight/volume). 25 mL of extract was analyzed by Kjeldahl for the NH_4^+-N measurement. 25 mL of H_2O extract was mineralized with 1 Kjeldahl Tablets Titanium (Buchi, United Kingdom) and 8 mL of H_2SO_4 96% for analysis ISO (PanReac AppliChem ITW Reagents) for the TKN. Table 2 shows the main characteristics of the digestate used for the experiment:

Table 2. Main characherististics of digestate. TN is Total Nitrogen (%); TKN is Total Kjeldahl Nitrogen (g/L); SOM is Soil Organic Matter and WC is Water Content. The results are the mean of 3 replicates for each sample.

Sample	**TN (%)**	**$\delta^{15}N$**	**NH_4^+-N (g/kg)**	**TKN (g/L)**	**SOM**	**WC**
Enriched zeo	0.40 ±0.02	13.98 ±0.61	-	-	-	-
Digestate	0.74	8.96	3.33 ±1.03	4.39 ±0.95	7.62	92.38

3.4.3 Agri-Food products

Together with leaves, also olives were collected (approximately 30-40 per plant). All olives have been pitted, dried at 60°C for three days and grounded. 2.5 g of the sample were subjected to acid digestion by the addition of HNO_3 (65% in distilled water, Supra-pur®, Merck KGaA, Darmstadt, Germany) and H_2O_2 (37% in distilled water, Suprapur®, Merck). In the same 50-mL Teflon digestion vessels, 3 mL of HNO_3 and 3 mL of H_2O_2 were added and the mixture was heated at 180°C until complete drying. Then, 3 mL of HNO_3 and 2 mL of H_2O_2 were added to the dry residue and it was heated until complete drying. The dry residue was resuspended in 2 mL of HNO_3 diluted to 100 mL with highly purified MilliQ® water before analysis by ICP-MS.

For the preparation of olive oil samples, 4 mL of MeOH were added to 3.5 mg of olive oil samples. Later, 10 µL of the previous stock was put with 1.1 mL of Fatty Acid Methyl Esters (FAME) and 0.3 mL of 19:0 standard; the solution was mixed in a Vortex for 10 seconds. 0.5 mL of MeOH-HCl were added to the solution and mixed in a Vortex for 10 sec. The mixture was heated for 60 minutes at 55°C. 2 mL of HQ-NaOH and 2 mL of Hexan were added and mixed twice for 10 sec in a Vortex. The samples were centrifugated for 2 minutes at 2000 rpm and 2 mL of the upper level of the solutions were transferred in clean vials. This step was repeated twice, adding 2 mL of Hexan in order to uptake as many amounts of the sample as we could. The final solutions were dried with N_2 at 40°C. They were resuspended in 100 µL of Isoctane and mixed in a Vortex for 10 sec. The samples were transferred in GC vials in order to quantify the Fatty Acids (FA) and to analyze the isotopic signature of FA (IRMS).

Chemical indices

The index of Bioaccumulation (BA) and the Translocation coefficient (TC) were measured in the olive tree and grape experiments in order to evaluate the ability of plants to uptake chemical elements from soil and transfer them to the fruit, respectively.

The BA was calculated as follows:

$$Index\ of\ Bioaccumulation\ (BA) = {C_{leaf}}/{C_{soil}} \qquad (2)$$

Where C is the concentration of a given element in leaf and soil where the plant is grown (Buscaroli, 2017). The TC was calculated as the concentration of an element in an aboveground biomass of the plant and the same concentration in another part (Alagić et al., 2015; Buscaroli, 2017; Vystavna et al., 2015). In other words, TC provides how each element is absorbed in different parts of the plant, leaf and olive in this study:

$$Translocation\ Coefficient\ (TC) = {C_{solid\ residue}}/{C_{leaf}} \qquad (3)$$

Where C is the concentration of a given element in the solid residue (olive) and in the leaf.

Crop Water Stress Index (CWSI) was calculated for leaves from the grapevine, in order to evaluate the water stress on plants. This parameter allows to normalize the temperature data with environmental conditions and to compare data during the time. Accordingly to Poblete-Echeverría and Zuñiga (2017), CWSI was calculated as follows:

$$CWSI = \frac{T_{canopy} - T_{wet}}{T_{dry} - T_{wet}} \qquad (4)$$

Where Tcanopy is the leaf temperature measured by a thermic sensor, T_{wet} means leaf temperature with complete transpiration (open stomata) and T_{dry} is leaf temperature with no transpiration (closed stomata). T_{wet} and T_{dry} can be calculated by empiric or experimental approaches. The CWSI coefficient can have values ranging from 0 to 1: higher values correspond to a water stress condition for the plant.

Also soil humidity and gas exchanges were measured by FDR mod. WaterScout SM 100 from Spectrum Technologies, Inc. and CIRAS2 Portable Photosynthesis System PP System – Amesbury, MA 01913 U.S.A. sensors, respectively.

3.4.4 Statistical analyses

All data were elaborated with R Studio 4.1.1 version.

To address significant differences between the treatments due to the zeolite application about new sustainable agriculture practices and Agri-Food traceability, parametric and non-parametric tests were applied. Normality and homoscedasticity were tested through Shapiro-Wilk and Barlett tests (p=0.05) for each variable. Data following normal distribution and with homogeneous variance were tested with a 1-way ANOVA and multiple comparison test (Tukey HSD) at p=0.05. If normality or homoscedasticity were not reached (even after log or ln transformation), a non-parametric test (Kruskal-Wallis) was applied instead of ANOVA.

Furthermore, the Pearson correlation method was applied for those elements with the highest chemical indices (BA and TC indices in Agri-Food traceability) in order to correlate the given element in soil, leaf and olive.

Principal Component Analysis (PCA) was applied to discriminate the groups of samples depending on the treatment variable (ZEO or CNT) or geographical origin, reducing the dimensionality of the dataset with a small loss of information. PCA is a mathematical procedure that transforms a number of possibly correlated variables into a smaller number of uncorrelated variables through a process known as orthogonal transformation (Dinov, 2018). In the simplest situation with n-observations $\{p_1, p_2, \ldots, p_n\}$ and two features $p_i = (x_i, y_i)$, a plot on the x-axis and y-axis can be drawn to represent them. With a personal coordinate system based on principal components, there is the first PC (PC1) which has the minimum distance fit in the feature space and the second PC (PC2) with a minimum distance fit to a line perpendicular to the first PC (Figure 6). In the same way, the third PC (PC3) would be a minimum distance fit to all previous PCs. Generally, in a 2D space, two PCs are drawn, which means the two PCs that represent the direction of the data that explain the maximal amount of variance. In hypothetical higher dimensional spaces, all PCs are needed to be figured. As a whole, PCA transformation preserves in its first PC as much variance in the original data as possible (Ruiz-Perez et al., 2020).

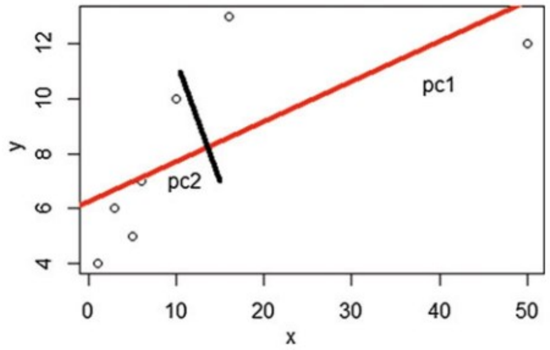

Figure 6. Schematic representation of the first two principal components (Dinov, 2018).

As described by Dinov (2018), the general formula for the PCs is:

$$PC_k = \sum_{i=1}^{N} a_{i,k} X_i \qquad (5)$$

Where k is any PC (PC1, PC2, …, PC_k), N represents the features, $a_k=\{a_{1,k}, a_{2,k}, \ldots, a_{N,k}\}$ maximize the variance of PC_k and X_i is a *n x 1* vector representing a column of the matrix X (complete design matrix with a total of n observations and N features). In conclusion, PCA has been used for discrimination for a long time because it is conducted on the original database and extracts a subset of scores (components). When the groups are well separated in score plots, it is possible to classify unknown data into the group with the closest average score. However, PCA is only capable of identifying gross variability and is not capable of distinguishing `among-groups' and `within-groups' variability (Barker and Rayens, 2003).

The Partial Least Squares Discriminant Analysis (PLS-DA) was applied to improve discrimination among groups according to the study area and the agricultural practices in the traceability topic. The Variable Importance in the Projection (VIP) indicates the separation power of each element: values higher than 1 (or near to this value) were considered as potential discriminators in the model.

PLS-DA is a multivariate dimensionality-reduction tool and it can be considered a “supervised” version of PCA. For these reasons, it can be used for feature selection and for classification. Generally, PLS-DA preserves in its first PC as much covariance as possible between the original data and its labeling (Ruiz-Perez et al., 2020). VIP scores summarise the influence of individual X variables on the PLS model and it is calculated as the weighted sum of squares of the PLS weight. It means that VIP scores give a measure useful to select what are the variables which contribute the most to the y variance explanation (Farrés et al., 2015).

The following packages were used for data analyses and figures in R studio: “ggplot2”, “Agricolae”, “Ggally”, “ggbiplot”, “ggfortify”, “corrplot” and “ggpubr” (Alboukadel Kassambara, 2020; Barret Schloerke, Di Cook, Joseph Larmarange, Francois Briatte and Marbach, Edwin Thoen, 2021; De Mendiburu, 2014; Masaaki Horikoshi and Yuan Tang, 2016; Taiyun Wei and Viliam Simko, 2021; Vincent Q. Vu, 2011; Wickham, 2016).

4. Results and Discussions of olive growing experiment

4.1 The effects of zeolitite on soil N dynamics and plant uptake

Soils from BN and BG experimental sites were characterized mainly by silty-clay-loam texture, with a slightly higher silty fraction in BG (Figure 7). On the other hand, the SL soils were characterized by an important sand fraction and were classified as sandy-loam and sandy-clay loam (Figure 7).

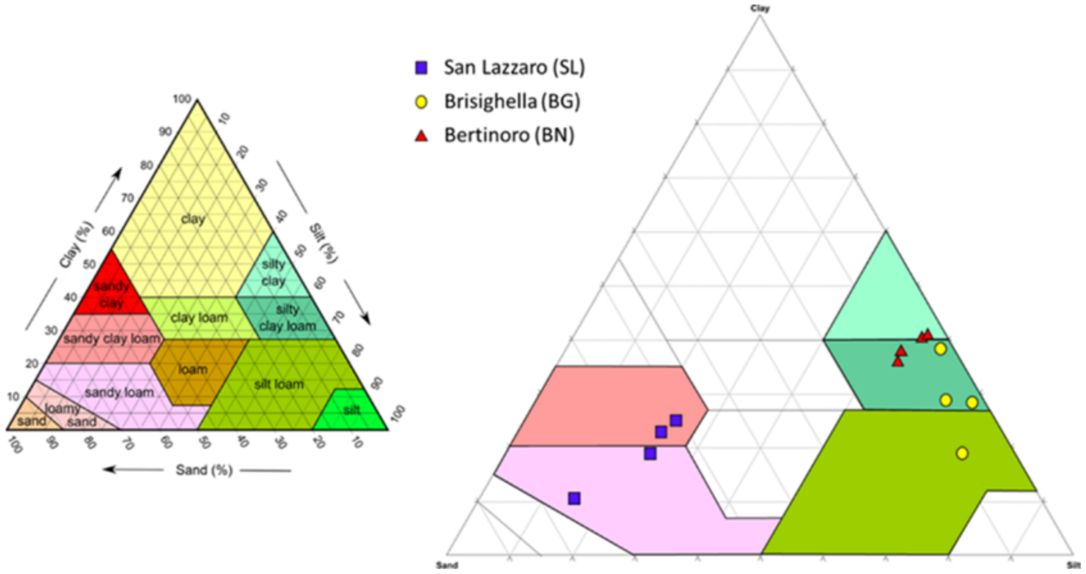

Figure 7. Particle size analysis and textural classification (USDA) of the soil samples from SL, BG and BN experimental fields.

Given the large dataset, in this work, we are discussing only annual trends and 3-years average significant observation. The complete dataset (average+st.dev of each variable at each sampling time and location is available in the Appendix).

In Table 3, the 3-year average of soil pH and SOM at each experimental site are reported. These basic parameters are indicators of soil quality and plant growth: SOM is the primary source of essential nutrients (N, P and S) and influences bulk density, water retention and soil temperature, as well as biological activity, and buffers pH (Paul, 2007).

Table 3. pH and Soil Organic matter (SOM) at each site (San Lazzaro, SL; Brisighella, BG; Bertinoro, BN). Data are divided by time of sampling (Pre-Fertilization, Post-Fertilization and Harvest) and treatment (ZEO and CNT). Average values represent a 3-year average (3 replicate/treatment per sampling, 3 sampling x per year, 54 samples in total per site). Means in the same column followed by different letters are significantly different ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests. The complete dataset is shown in the Appendix.

		SL				BG				BN			
		pH		SOM (%)		pH		SOM (%)		pH		SOM (%)	
Pre-Fert	**CNT**	8.01a	±0.09	3.59a	±1.12	8.83a	±0.29	4.76ab	±1.32	8.84a	±1.21	6.44b	±0.03
	ZEO	7.93a	±0.37	4.04a	±0.48	8.77a	±0.22	5.51ab	±2.04	8.86a	±1.24	6.30b	±0.33
Post-Fert	**CNT**	7.71a	±0.43	5.05b	±0.80	8.66b	±0.19	5.26b	±0.30	8.65a	±0.92	6.97a	±0.38
	ZEO	8.27a	±0.55	3.18b	±2.72	8.67b	±0.08	5.22b	±0.94	8.45a	±0.96	7.64a	±0.63
Harvest	**CNT**	7.56a	±0.24	3.21c	±0.62	8.77ab	±0.05	5.91a	±0.51	8.47a	±1.11	5.35b	±2.37
	ZEO	7.56a	±0.48	3.02c	±0.51	8.84ab	±0.13	6.16a	±0.25	8.43a	±1.09	4.79b	±3.32

In the BG site, a slight decline in soil pH was observed in Post-Fert samples (Table 3), probably due to chemical fertilizer addition. In BG and BN, SOM is generally higher than in SL site, due to the different soil texture. The presence of silt and clay in fact maintains more C from primary production and increases SOM under certain environments (Eldor A. P., 2007). SOM could be influenced by fertilization and irrigation of soil, and they are correlated with SO_4^{2-}, PO_4^{3-} (Figure 8), $\delta^{15}N$ (Figure 9) and Total Carbon (TC) (Figure 10). However, in SL and BN, SOM was higher in Post-Fert than in Pre-Fert and Harvest, while BG showed an opposite trend (Table 3). Moreover, Total Carbon (TC) in BG and BN sites confirmed the higher trend of SOM explained above, while SL showed an opposite trend (Figure 10). As far as pH and SOM are concerned, no significant effects related to the zeolitite addition to soil were observed over the 3 years of experimentation. The nutrient input reduction of 50% every year in ZEO treatment suggests a more favorable balance between inputs and outputs of SOM in the zeolitite-added soils.

Due to the different kind of fertilizer used, the SL site showed SO_4^{2-} and PO_4^{3-} values remarkably higher than BG and BN fields. The chemical fertilizer applied in SL in fact contained phosphate and sulfate, unlike the fertilizers used in BG and BN. Being both SO_4^{2-} and PO_4^{3-} negatively charged, they are unsuitable for cation exchange by natural zeolites, which led to non-significant differences in the retention of these ions in the soil between CNT and ZEO. However, given the lower amount of fertilizers applied to ZEO, a lower values of SO_4^{2-} and PO_4^{3-} were expected in this treatment, at least after fertilizer application. SL highlighted its highest values Post-Fertilization (Figures 8C and D), while BG and BN values showed no differences during the agronomic year.

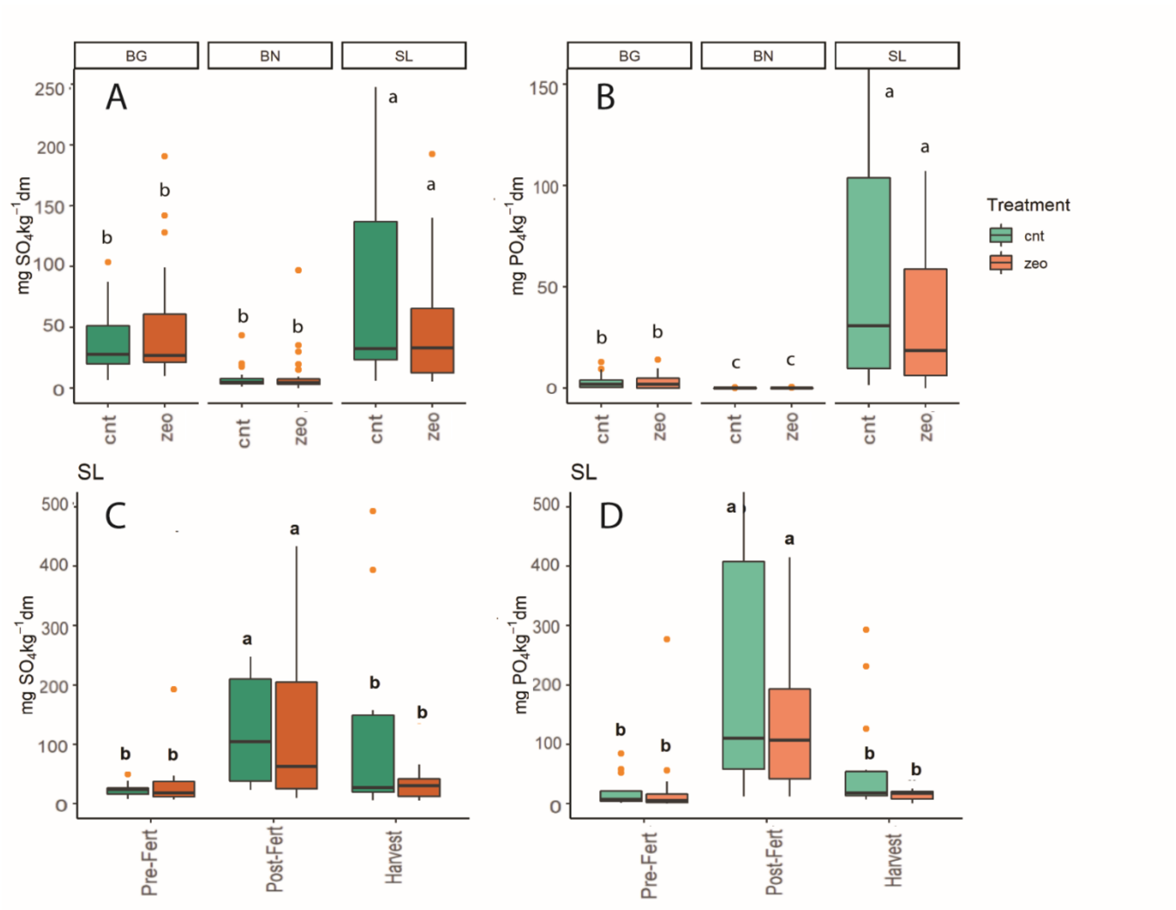

Figure 8. Box-plot of SO$_4^{2-}$ (**A**), PO$_4^{3-}$ (**B**) content of soil samples. The graphs are divided by experimental site (BG, BN, SL) and treatment (CNT and ZEO). Box-plot of SO$_4^{2-}$ (**C**), PO$_4^{3-}$ (**D**) content of soil samples from SL site. The graphs are divided by sampling (Pre-Fert, Post-Fert and Harvest) and Treatment (ZEO and CNT). Figure 8A,B: The graphs are constructed considering a 3-year average based on 27 samples per treatment at each site. Figure 8C,D: The graphs are constructed considering the site specific 3-year average (9 observations at each sampling time per each treatment, 54 total observations). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

The different kind of chemical fertilizers adopted in the experimental sites also influenced the N isotopic composition in the soil, as clearly shown in Figure 9A. On average, the δ^{15}N of SL soil is higher than in the other sites due to the use of organic fertilizers, which generally have higher ^{15}N content than the synthetic ones (Bateman and Kelly, 2007), but no significant variation over the agronomic year occurred (Figure 9B). At the BG site, after the addition of chemical fertilizers the δ^{15}N of soil tended to decrease (Figure 9C) while at the BN site, no differences were detected (Figure 9D). In natural ecosystems, soil δ^{15}N ranges from −6‰ to 16‰ (Shan et al., 2019) and this high variability can be related to climate gradients and different atmospheric conditions. An inverse and a direct correlation between mean annual precipitation (MAP) and mean annual temperature (MAT) can be in fact observed with δ^{15}N (Amundson et al., 2003). BG presents the highest MAP, while both BG and BN sites show the lowest MAT during 2019–2021. An increase in the δ^{15}N values at BG is also observed in concomitance with the harvest (Figure 9C), probably due to the temperature peaks recorded during the summers of 2020 and 2021. The lower $\delta^{15}N$ values of BG and BN with respect to SL could thus result both from different N sources and climatic conditions.

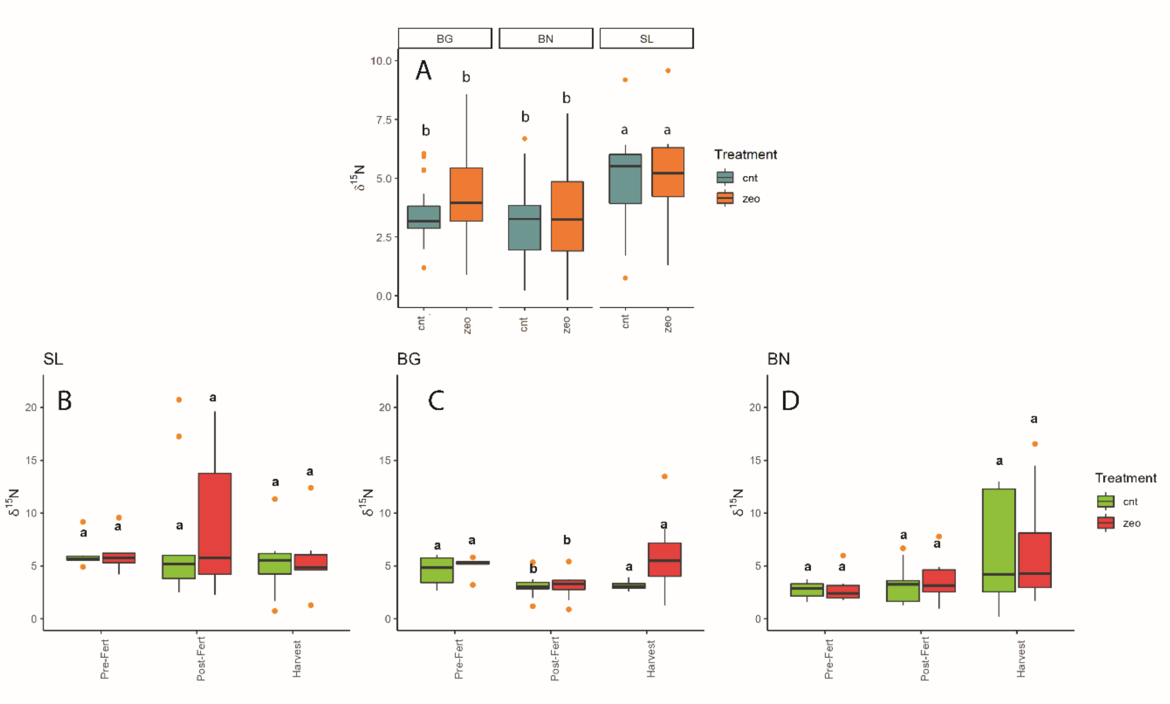

Figure 9. Box-plot of $\delta^{15}N$ (**A**) of soil samples of the three sites. The graph is divided by the experimental site (BG, BN, SL) and treatment. Boxplots of $\delta^{15}N$ of San Lazzaro (SL) (**B**), Brisighella (BG) (**C**) and Bertinoro (BN) (**D**). The graphs are divided by agronomic season (Pre-Fert, Post-Fert and Harvest) and treatment (CNT and ZEO). Figure A: the graph is constructed considering a 3-year average based on 27 samples per treatment at each site. Figure 9B–D: the graphs are constructed considering the site specific 3-year average (9 observations at each sampling time per each treatment, 54 total observations). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

To evaluate the influence of irrigation, the Carbon Discrimination Factor ($\Delta^{13}C$) was calculated from $\delta^{13}C$ data. Figure 11 shows the $\Delta^{13}C$ for leaves of each site, which means the $\delta^{13}C$ normalized for changes in atmospheric CO_2 concentration through Equation (1), where a and p refer to air and plant (Farquhar et al., 1989).

$$\Delta = \frac{\partial a - \partial p}{1 + \partial p} \tag{5}$$

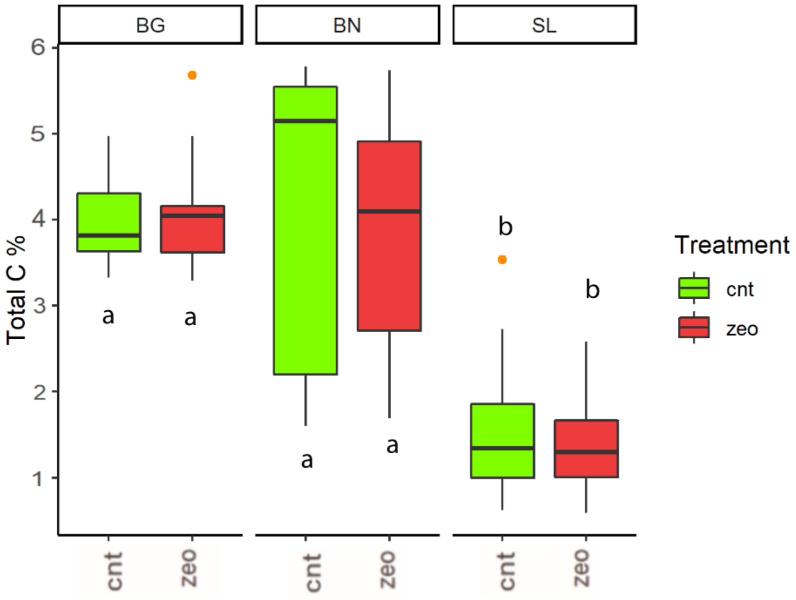

Figure 10. Box-plot of Total Carbon (TC) of soil samples in San Lazzaro (SL), Brisighella (BG) and Bertinoro (BN). The graphs are divided by treatment (CNT and ZEO). Data are the results of 3 years of experiment: For each year, 3 samplings with 3 replicates per treatment were sampled (54 samples per each site, divided in 27 samples per treatment). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

According to Riehl et al. (2014) a 1‰ $\Delta^{13}C$ variation can be used to distinguish stressed from well-watered plants without accounting for soil fertility effects. Water stress conditions in fact causes a decrease in photosynthesis, transpiration and leaf conductance which in turn modify the carbon isotopic composition (Busch et al., 2020; Farquhar et al., 1989; Kumar and Singh, 2009). As it is known, zeolites can adsorb water molecules in their structure, which means an increase in the overall soil water holding capacity and the consequent possibility to reduce irrigation (Jakkula V., 2018). Nevertheless, no significant variations between ZEO and CNT treatments were highlighted by the $\Delta^{13}C$ data. This fact is partially in contrast with the results obtained by Ferretti et al., (2017a) where a change in $\Delta^{13}C$ in maize and wheat grown in soil amended with the same natural zeolite- rich tuff was observed. Although in that case, the authors ascribed the $\Delta^{13}C$ variations to the manuring effect. In our case, a significant difference in $\Delta^{13}C$ was observed in BG only, likely due to the additional water provided to the plants' trough irrigation. This site in fact is the only one which underwent artificial irrigation, added to the highest MAP over the three years of experimentation.

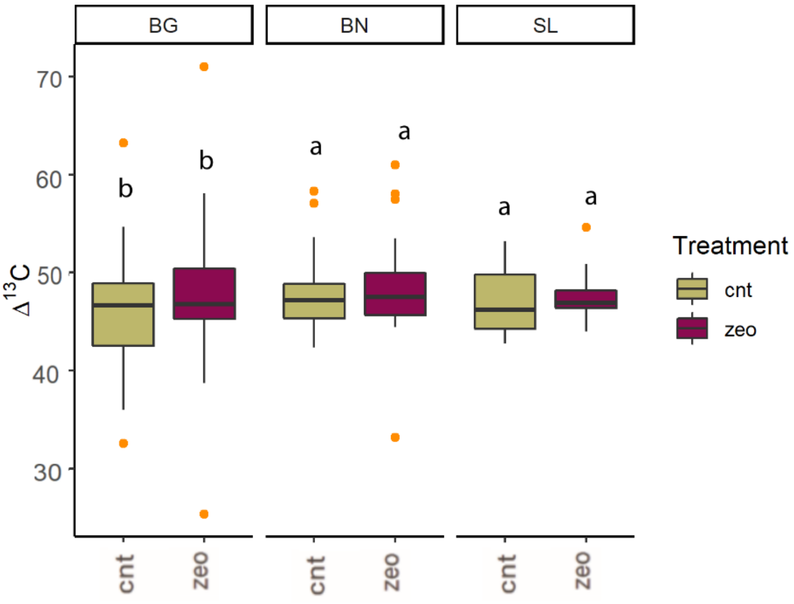

Figure 11. Box-plot of $\Delta^{13}C$ of leaves divided by area and treatment. The figure is constructed considering a 3-year average based on 27 samples per treatment at each site. Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

N is one of the most important nutrients for plants. Thus, analyses of its different inorganic speciation were performed to address the effects of natural zeolites on soil N cycling in the three experimental fields. Nitrite (NO_2^--N) usually does not accumulate in soils because it is an intermediate product of nitrification (that transforms NH_4^+ into NO_3^--N), or it is denitrified to NO and N_2O and N_2 gases. On the other hand, nitrate (NO_3^--N) is one of the main forms of N used by plants and can also be exploited by microbes to satisfy their N needs (immobilization processes) (Hatfield and Follet, 2018). Nitrate, however, can follow various transformation pathways which may also lead to N losses in the atmosphere (as nitrous oxides due to incomplete denitrification) and/or can be leached into the water system as a result of anionic repulsion by soil particles.

The results of TN analyses of soils and leaves and NO_3^--N and NO_2^--N of soils are shown in Figures 12-14 for SL, BG and BN sites, respectively (3-year average).

In SL no differences were observed between the different treatments, although the N input in CNT treatment was twice that in ZEO treatment. NO_2^--N of soil (Figure 12A) did not show any difference between treatments (ZEO and CNT) or sampling time (Pre-Fert, Post-Fert and Harvest) among the 3 years of the project, showing values always below 10 mg kg^{-1}. Even NO_3^--N in soil samples (Figure 12B) showed no significant variations among the treatments. A remarkable difference between CNT treatment Post-Fertilization and Harvest can be observed, probably due to NO_3^--N removing processes (gaseous losses, leaching, microbial immobilization or Dissimilatory Nitrate Reduction to Ammonium). This evidence is partially sustained by a tendency to a lower N storage in olive leaves at the Harvest in the CNT (although not significant). Soil TN (Figure 12C), reflects the same trend for nitrate, showing no significant differences between treatments. The seasonal fluctuations of these N species (with higher values after fertilization) are related to the input of N brought by fertilizers. The TN of leaves (Figure 12D) likely supports this hypothesis because the leaves have shown no differences in N content due to treatments or time. However, they showed an opposite trend to that of soils, due to the different availability of N during the agronomic year in different environmental compartments. Immediately after fertilization, TN is concentrated in the soil, and it is lower in leaves while at the harvest the trend was opposite.

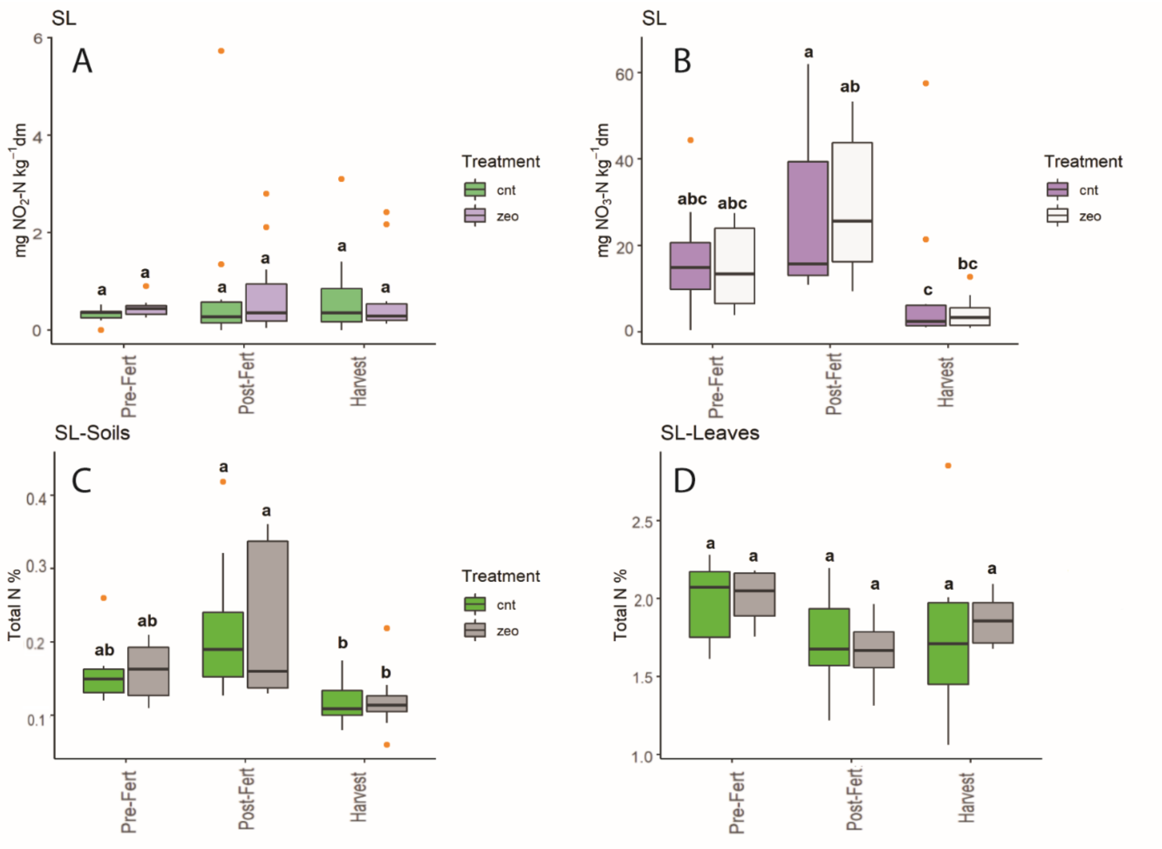

Figure 12. Box-plot of NO_2^--N (**A**), NO_3^--N (**B**) content of soil samples, Total Nitrogen (TN) of soils (**C**) and leaves (**D**) in San Lazzaro field (SL). Graphs consider the site specific 3-year average (9 observations at each sampling time per each treatment, 54 total observations). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

In the BG site, TN did not show any significant difference due to the treatment and sampling time for both soils (Figure 13C) and leaves (Figure 13D), coherently to the SL site. The NO_2^--N (Figure 13A) and NO_3^--N (Figure 13B) of BG soils showed a trend similar to SL and no differences were accounted for between ZEO (50% of fertilizer) and CNT (100% of fertilizer). However, sampling time significantly affected the amounts of N in the soil. NO_2^--N (Figure 13A) in Post-Fertilization ZEO samples showed significant differences with respect to ZEO at Harvest, suggesting lower nitrite production (or improved consumption) in this treatment. The NO_3^--N (Figure 13B) content in CNT treatment at Harvest was significantly lower than Post-Fertilization, but an increase in N uptake of plants is not able to explain the NO_3^--N reduction in soil. This decrease is probably due to a N loss in the surrounding environment which did not happen for ZEO treatments, as suggested by the tendency of ZEO leaves to have higher TN amounts for all sampling stages, although not statistically significant.

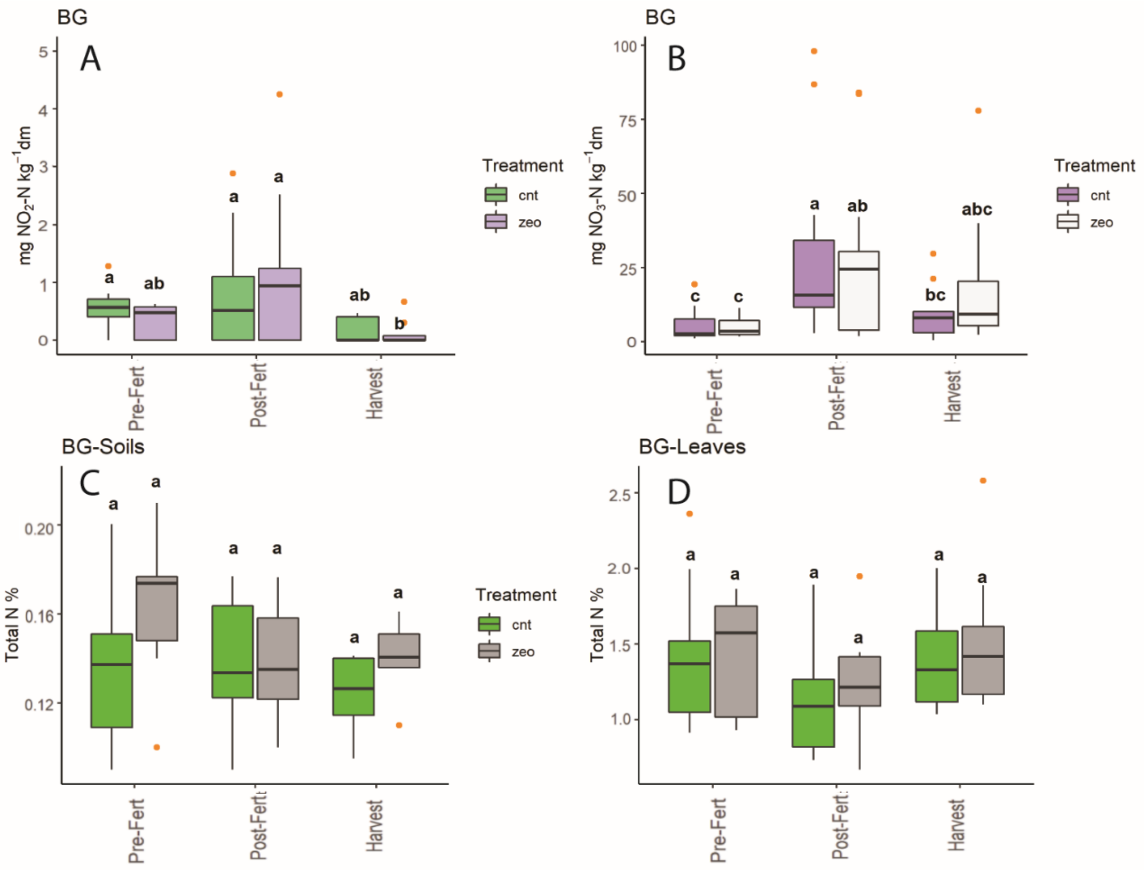

Figure 13. Box-plot of NO_2^--N (**A**) and NO_3^--N (**B**) content of soil samples, Total Nitrogen (TN) of soils (**C**) and leaves (**D**) in Brisighella field (BG). The graphs are constructed considering the site specific 3-year average (9 observations at each sampling time per each treatment, 54 total observations). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

The BN samples showed a trend similar to the SL and BG areas during the three years of monitoring. NO_2^--N (Figure 14A) and NO_3^--N (Figure 14B) of soils showed no significant differences between ZEO and CNT. NO_2^--N showed significant differences between Pre-Fertilization and Post-Fertilization samplings, with higher values at Pre-Fert. NO_3^--N followed the trend linked to the fertilization, with higher values at Post-Fertilization right after the N input. The ZEO treatment in Pre-fertilization is similar to the CNT in Post-Fertilization (where twice the amount of fertilizer was applied with respect to the ZEO treatment), indicating that zeolite probably helped the soil to retain more N available to the plant during time. TN of soils (Figure 14C) revealed no variations due to the treatments or sampling time, and no other differences were highlighted neither for TN of leaves (Figure 14D) nor for the SL and BG sites. As for BG, also in BN a tendency for a higher N content of leaves was recorded although not significant from a statistical point of view.

In general, the results of N dynamics over the 3 years of monitoring in the 3 experimental sites indicate that notwithstanding 50% fewer N inputs, the soil N content was similar between CNT and ZEO. Given that no differences in N uptake by plants were observed, this evidence leads to the hypothesis that zeolite minerals helped to reduce N losses and promoted N storage in the soil, augmenting the fertilization efficiency.

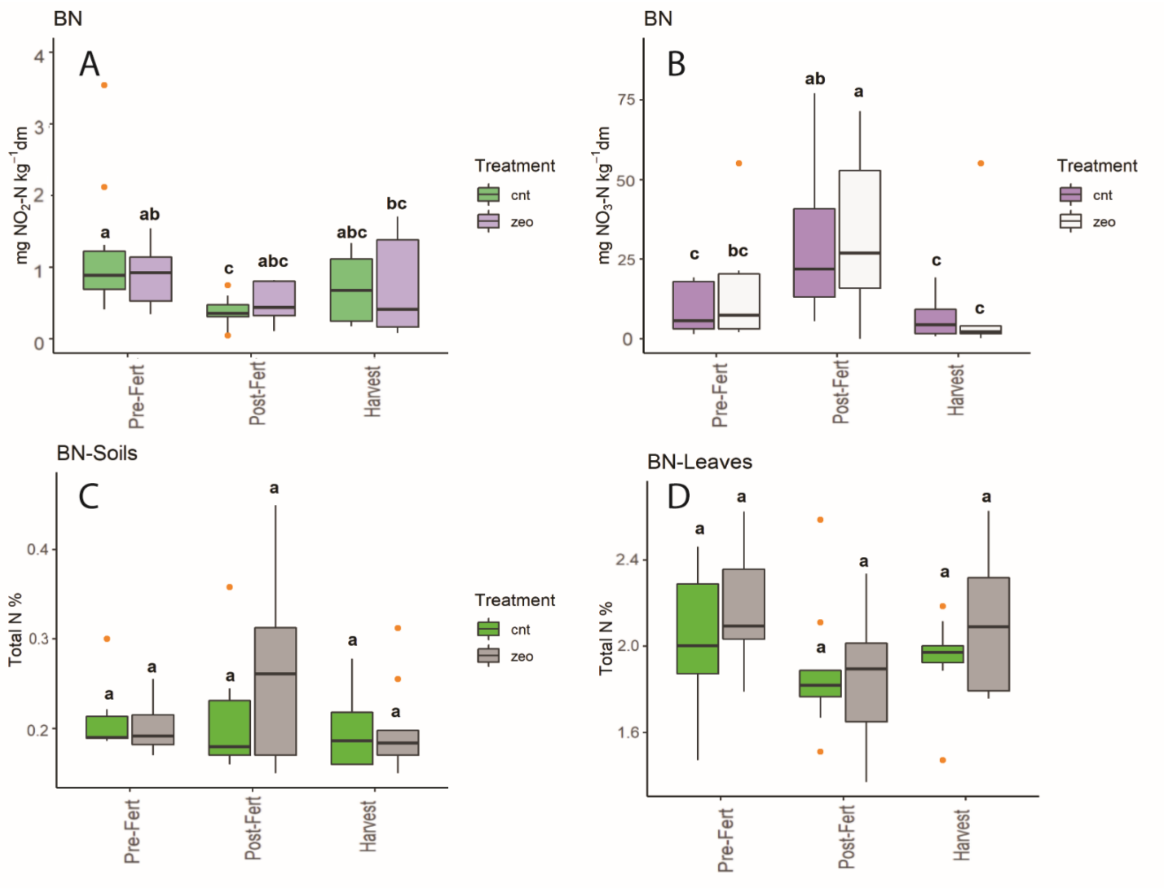

Figure 14. Box-plot of NO_2^--N (**A**) and NO_3^--N (**B**) content of soil samples, Total Nitrogen (TN) of soils (**C**) and leaves (**D**) in Bertinoro field (BN). The graphs are constructed considering the site specific 3-year average (9 observations at each sampling time per each treatment, 54 total observations). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

The analysis of variance of the data collected in the BG field did not reveal any difference between the two treatments (Table 4), while some differences between ZEO and CNT were highlighted in both SL and BN fields. In the SL site, tree height, number and length of shoots were higher for Cv

Montebudello and *Farneto* for ZEO treatment than for CNT (Table 5). The number of shoots was greater in the ZEO thesis for Cv *Colombina*, while other measurements exhibited no significant difference compared to CNT. The Cv. *Capolga* in the BN field showed no differences in the level of growth of the aerial part, while in the other two cultivars (*Colombina* and *Correggiolo*), a significantly greater development in the plants treated with natural zeolite-rich tuffs was observed, despite the reduced dose of fertilizer applied (Table 6).

Table 4. Vegetative measurements of olive trees grown on soil treated with natural zeolitite (ZEO) versus plants grown on unamended soil (CNT). Data are expressed as a mean of 15 replicates per thesis.

		Tree Height (cm)	**Number of Branches**	**Average Branches Length (cm)**	**∑ Branches Length (cm)**
Cv. *Nostrana di Brisighella*	**ZEO**	141.48	63.39	30.63	1918.05
	CNT	134.09	51.78	30.09	1597.65

Table 5. Vegetative measurements of olive trees grown on soil treated with natural zeolitite (ZEO) versus plants grown on unamended soil (CNT). Data are expressed as a mean of 15 replicates per thesis. The bold font indicates statistically significant differences between the groups ($p < 0.05$).

		Tree Height (cm)	**Number of Branches**	**Average Branches Length (cm)**	**∑ Branches Length (cm)**
Cv. *Montebudello*	**ZEO**	**121.14**	**49.86**	24.71	**1193.14**
	CNT	**92.00**	**20.00**	22.39	**441.86**
Cv. *Farneto*	**ZEO**	**114.31**	**69.69**	22.75	**1592.50**
	CNT	**84.54**	**44.38**	20.37	**972.15**
Cv. *Montecapra*	**ZEO**	104.50	**62.63**	22.71	**1368.38**
	CNT	99.29	**47.00**	21.55	1060.50

Table 6. Vegetative measurements of olive trees grown on soil treated with natural zeolitite (ZEO) versus plants grown on unamended soil (CNT). Data are expressed as a mean of 15 replicates per thesis. The bold font indicates statistically significant differences between the groups ($p < 0.05$).

		Tree Height (cm)	**Number of Branches**	**Average Branches Length (cm)**	**∑ Branches Length (cm)**
Cv. *Capolga*	**ZEO**	86	20.76	14.65	392.18
	CNT	83.1	18.95	13.92	347.18
Cv. *Colombina*	**ZEO**	74.88	**10.53**	**15.63**	**233.82**
	CNT	63.77	**6.46**	**11.33**	**141.58**
Cv. *Correggiolo*	**ZEO**	**102.38**	9.25	**21.04**	**291.38**
	CNT	**80.43**	7.79	**15.76**	**204.39**

The only field where no differences were observed between the two treatments is BG, the irrigated field. It is possible that the action of the zeolite, in addition to reducing N leaching and increasing the Nitrogen Use Efficiency (NUE), takes place at the water level (although no differences were observed by Δ^{13}C), so in orchards without any water deficits, it is harder to account for differences in plant development.

These results are in agreement with those of Prisa (2020), that found an increase in agronomic characteristics in plants of *Ranunculus asiaticus* treated with zeolites, and with Choo et al. (2020) that found an increased number of fruits and greater fruit yield in papaya plants treated with zeolites.

To evaluate the benefits of using zeolitite in olive growing, the general comparison of treatments year by year is presented in this chapter. pH, SOM and TC of soils are shown in Figure 15, while in Figure 16 each of the investigated N species is shown.

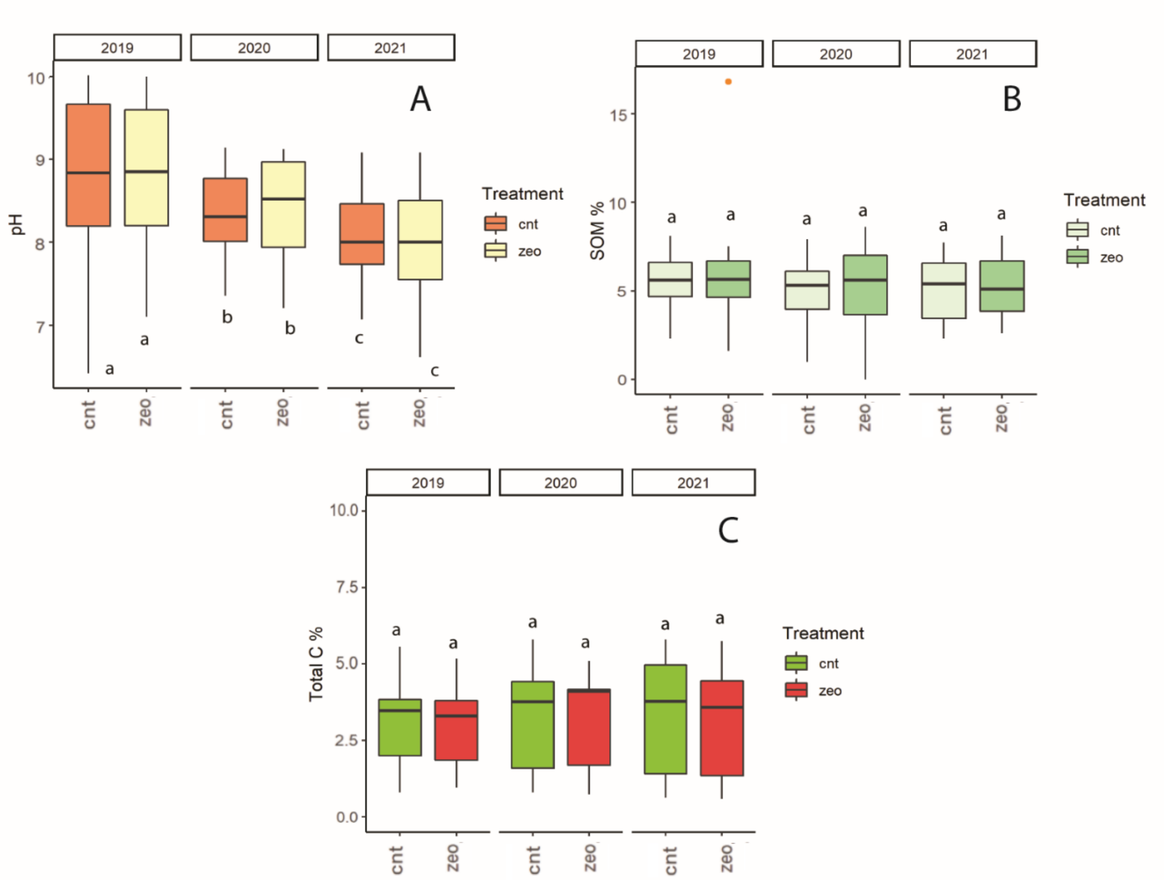

Figure 15. pH, Soil Organic Matter (SOM) and Total Carbon (TC) are shown for the three years of the project. Data are divided by year (2019, 2020 and 2021) and treatment (with zeolite and control for (**A**) pH, (**B**) Soil Organic Matter (SOM) and (**C**) Total Carbon (TC). The graphs are constructed considering the year specific average for all three sites (27 observations at each year per each treatment, 54 total observations per year). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

In all experimental fields, pH did not undergo any significant difference between CNT and ZEO treatments. A global trend towards acidification of the soil from 2019 to 2021 can be however observed (Figure 15A), which can be a consequence of the leaching of exchangeable bases such as Mg^{2+}, K^+ and Ca^{2+} because of intense precipitation or irrigation practices. No significant differences were accounted between SOM and TC and they did not change in relation to different treatments and during time (Figures 15B, C). Although SOM and TC could be influenced by fertilization and irrigation practices, we did not observe any significant variation. At the same time, SOM, as well as TC, did not decrease over the 3 years of experimentation, proving that the use of zeolitite did not influence these parameters in soil but helps preserving SOM even with a reduced amount of nutrient inputs while maintaining or even improving the plant development.

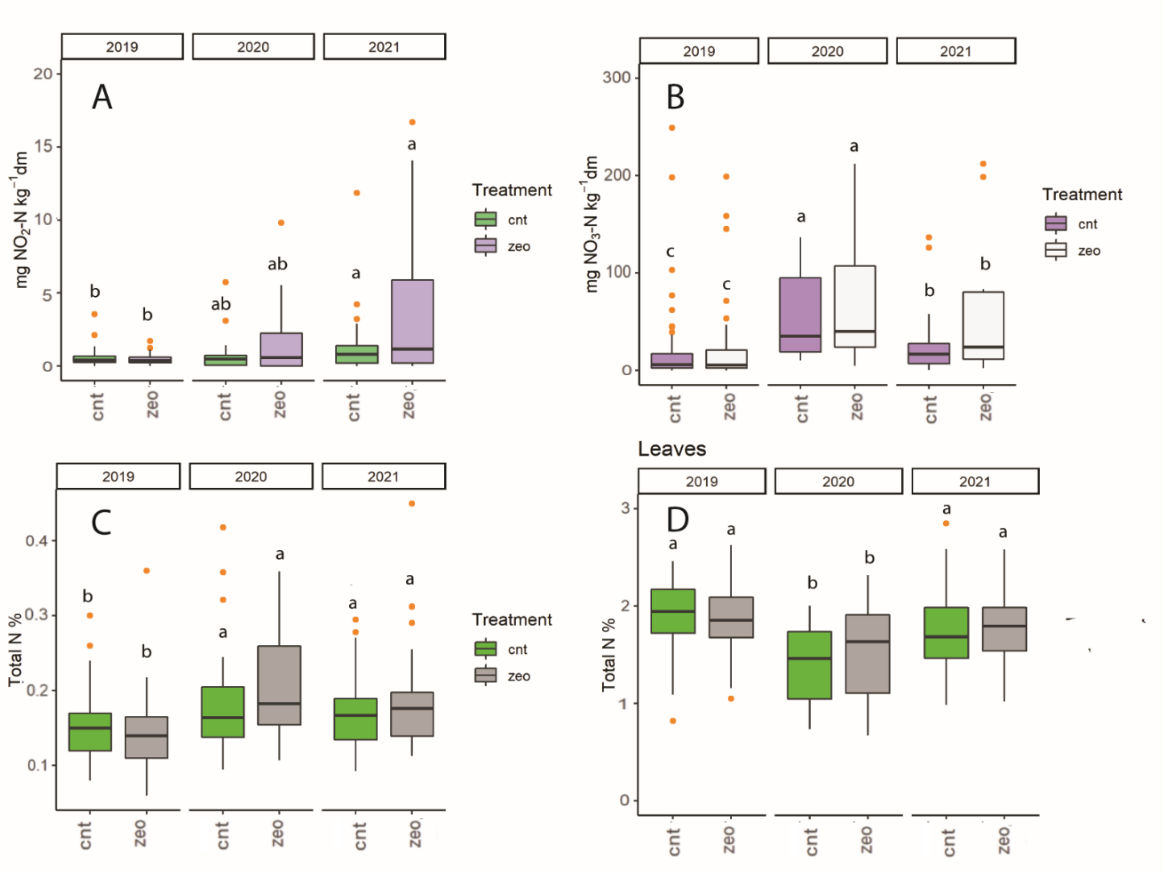

Figure 16. Box-plot of NO_2^--N (**A**) and NO_3^--N (**B**) content of soil samples, Total Nitrogen (TN) of soils (**C**) and leaves (**D**) divided by years and treatment. The graphs are constructed considering the year specific average for all three sites (27 observations at each year per each treatment, 54 total observations per year). Different letters represent significant differences ($p < 0.05$) as a result of ANOVA and Tukey (HSD) tests.

The only difference that occurred in nitrogen species was linked to the time and to the type and amount of fertilizer applied to each field: (1) NO_2^--N showed a significant difference among years, with concentrations that increased from 2019 to 2021; (2) NO_3^--N in soil was significantly different during 3 years, with the lower values recorded in 2019 and the higher values recorded in 2020 and 2021; (3) TN in soil showed a very similar pattern to that of NO_3^--N with an increase after the first experimental year (2019) and (4) The TN of leaves was lower in 2020 (opposite trend to the NO_3^--N). For each N species, no differences were accounted for between CNT and ZEO treatments (notwithstanding the 50% reduction of fertilizers), as already demonstrated in detail for each experimental site.

TN of leaves strongly indicates that plants did not uptake more N in CNT than in ZEO treatment, although ZEO leaves showed a slight tendency in higher N uptake in 2020 and 2021 (not statistically significant), which can be caused by an augmented availability of N among the years.

Principal Component Analysis (PCA), which is often used to discriminate the groups of samples, reducing the dimensionality of the dataset without a large loss of information, was applied only to the data related to the ZEO and CNT treatments during the three years of the project.

PC1 and PC2 axes explained 48.59% of the total variance, divided into 30.61% of the First Principal Component (PC1) and 17.98% of the Second Principal Component (PC2). All the data showed a positive correlation in PC1, except Soil Organic Matter (SOM), Total Carbon (TC) and pH. Instead, in PC2 only Carbon Discrimination Factor ($\Delta^{13}C$) had a positive correlation, while all other parameters highlighted a negative correlation with PC2. This low value of total variance does not allow for distinguishing between the different treatments, thus further supporting the hypothesis that CNT and ZEO treatments were not different, notwithstanding the fertilizer input reduction of 50% in the ZEO treatment.

The similar N uptake recorded by the leaves in the three different experimental sites, as well as the tendency for a better development of plants grown on zeolite-amended soil, notwithstanding the 50% N input reduction, strongly suggest that in CNT treatment larger N losses occurred, leading to negative environmental and economic effects. On the other hand, the presence of zeolitite in the soil maintained the nutrient for a longer time contributing to a healthier condition for plants and yield production.

It is well known that zeolitite as a soil amendment reduces N leaching and increases Nitrogen Use Efficiency (NUE) and crop yield (Sepaskhah and Barzegar, 2010). Since the addition of zeolitite probably influenced several pathways of N losses, it also allowed a more sustainable use of N fertilizers. Furthermore, the N in the topsoil is strongly related to agricultural practices and is influenced by the amount and form of the fertilizers used. This N can be easily lost by leaching, NH_3 volatilization and other N gas losses. Chemical fertilizers, such as urea, can lose even more than 30% of the applied N as NH_3 in the few hours after the spreading, if the conditions for volatilization are met (Soares et al., 2012). Ferretti et al. (2017) found evidence of a higher FUE in zeolitite-amended soil after performing an isotopic tracing in the soil-plant system. In another study, it was demonstrated that in similar conditions, NH_3 emissions can be reduced up to 60% using the same type of zeolitite used in this work (Ferretti et al., 2017a). Consequently, the application of zeolitite to soil can be the key to reducing N losses in the environment, allowing a significant reduction in fertilizer N inputs (50%), maintaining or even increasing the vegetative development.

The mechanism through which zeolitite is able to maintain the nutrients in the soil for longer periods of time is, however, still a matter of debate. Ferretti et al. (2021) employed the ^{15}N pool dilution technique to measure gross N transformation rates in zeolitite-amended soil and found no evidence of increased ammonification in soil treated with natural zeolites in the short-term. Thus, the efficiency of zeolitites (at natural state) cannot be explained by an increased production of new mineral N from organic matter decomposition. However, from the same study emerges a slight "delay" effect on gross nitrification. Apparently, in zeolitite-amended soil, ammonium is more slowly converted into nitrate. Thus, the mechanism that might be responsible for the improved NUE in the treated soil is the perturbation of various abiotic parameters after the addition of zeolite minerals (CEC, water retention) that is reflected in different biotic processes in the short-term, probably altering the quantity of N available for plant uptake. In another short-term incubation study at laboratory scale, it was observed that the exchange of N between minerals and the surrounding environment is very fast. Thus, N is accessible to microbial biomass in the short-term but only mild effects on the microbial community (fungal/bacterial ratio) and on N transformation rates were observed (Ferretti et al., 2021; Sepaskhah and Barzegar, 2010). Thus, it is likely that the zeolites reduce the N mobility in the short-term and delay the transformation of ammonium into nitrate, resulting in "more time" for plants to uptake N and, by consequence, in a lower demand for N fertilizers and N losses.

4.2 Chemical analysis of soil, leaves and olives

Soil samples showed a variable geochemical fingerprint in terms of trace elements (Table 7). The highest values were recorded for Sr, followed by Rb, (values between 100-50 ppm), Pr, Nb, Ce, Sm and Zr (values between 50-15 ppm). All the other elements showed concentrations ranging between 10-0.15 ppm.

All leaf samples (Table 8) showed similar values in terms of Rb and Sr including between 12 and 1 ppm, whereas other elements showed higher variability due to the location (MN or SL) and/or applied treatments.

Rb, Tm and Lu elements showed significant differences ($p<0.05$) due to the geographical origin of leaves, with higher values for MN than SL site.

Concerning trace element concentrations in MN leaves, significant differences were found between the various treatments ($p<0.05$). Almost all elements were more concentrated in ZN-DM treatment than in DM and CNT, except for Rb and Sr. Nb, La, Ce and Pr were more concentrated in ZN-DM, followed by DM and then CNT with values around 1-0.5 ppm in ZN-DM and decreasing by a factor 10 and 100 in DM and CNT. Although all other elements showed values less than 500 ppb, ZN-DM showed the highest trend. The application of dimethoate thus increased the concentration of the elements in the leaf, but a stronger contribution was brought by the application of natural zeolites which increased their concentration by a factor of 200 compared to the CNT.

In SL leaves, Th and U (150-20 ppb), Zr, Nb, Ce (600-100 ppb), Pr and Sm (around 100 ppb), Tb and Hf (around 20 ppb) highlighted higher values in ZN and ZA treatments than in traditional SF. All other elements did not show any differences between treatments ($p>0.05$).

A comparison of our data with the literature is difficult because of the variability due to the treatment and geographical origin of our samples, but we can generally compare values of Rb (2-12 ppm) and Sr (38-55 ppm) with data of Pošćić et al., (2020) and Higueras et al., (2012). All other elements showed values lower than in the literature survey (Higueras et al., 2012; Pošćić et al., 2020). However, our results agree with the median REE concentrations in leaves (from 6 to 100000 ppb) studied by Pošćić et al., (2020).

Table 7. Trace elements composition (ppm) of soil samples (mean ± standard deviation). The values are the result of three replicates per area. MN and SL are Montiano and San Lazzaro sites. Different letters indicate significant difference ($p<0.05$) as a result of ANOVA and Tukey HSD tests.

	MN		**SL**	
REE	**mean**	**dev std**	**mean**	**dev std**
Rb	69.03a	±14.24	69.36a	±14.20
Sr	101.78a	±27.61	82.23b	±10.08
Zr	16.33a	±5.76	17.68b	±2.48
Nb	32.71a	±2.26	23.31b	±6.11
La	8.35a	±0.96	6.00a	±0.89
Ce	22.35a	±7.08	21.62a	±5.57
Pr	42.29a	±12.56	45.10a	±10.67
Nd	4.95a	±1.54	5.20a	±1.17
Sm	19.12a	±6.06	19.99a	±4.40
Eu	3.75a	±1.20	4.05a	±0.85
Gd	0.78a	±0.25	0.88a	±0.19
Tb	3.18a	±1.05	3.58a	±0.69
Dy	0.54a	±.0.18	0.62a	±0.11
Y	2.60a	±0.86	3.01a	±0.51
Ho	0.50a	±0.17	0.59a	±0.10
Er	1.36a	±0.45	1.58a	±0.27
Tm	0.21a	±0.07	0.25a	±0.05
Yb	1.28a	±0.42	1.48a	±0.34
Lu	0.18a	±0.06	0.22a	±0.04
Hf	0.91a	±0.07	0.76a	±0.21

Ta	0.67a	±0.13	0.76a	±0.37
Th	8.03a	±2.66	8.18a	±2.56
U	1.83a	±0.16	1.75a	±0.44

Table 8. Trace elements composition (ppm) of leaf samples (mean ± standard deviation). Values with "*" are expressed in ppb. The values are the result of three replicates. MN and SL are Montiano and San Lazzaro sites. In the MN field, CNT refers to the negative control, DM refer to Dimethoate treatment while ZN-DM refers to natural zeolite + Dimethoate treatment. In SL field, SF refers to the traditional treatment of Spinosad+Spyntor fly, ZA is referred to the NH4+-enriched zeolite and ZN is natural zeolite treatment. "nd" refers to not-detected value. Different uppercase letters indicate significant differences due to the geographical origin while lowercase letters indicate significant differences between the treatments within the same experimental site (p<0.05) as a result of ANOVA and Tukey's HSD test.

REE	MN						SL					
	DM		**CNT**		**ZN-DM**		**SF**		**ZN**		**ZA**	
	Mean	**dev std**	**mean**	**dev std**	**mean**	**dev std**	**mean**	**dev std**	**mean**	**dev std**	**mean**	**dev std**
Rb	10.32Aa	±1.60	6.64Aa	±2.99	11.45Aa	±2.36	6.45Ba	±1.03	4.67Ba	±0.63	2.33Bb	±0.60
Sr	54.67Aa	±2.30	40.55Aa	±17.79	52.40Aa	±12.86	44.10Aa	±11.41	38.72Aa	±1.69	39.99Aa	±2.89
Zr	0.12Ab	±0.01	0.08Ac	±0.03	0.20Aa	±0.04	0.12Ab	±0.10	0.14Aa	±0.03	0.11Aa	±0.01
Nb	0.14Ab	±0.01	0.04Ac	±0.02	1.28Aa	±0.00	0.04Ab	±0.00	0.42Aa	±0.04	0.62Aa	±0.19
La	0.013Ab	±0.003	0.004Ac	±0.003	0.103Aa	±0.018	0.004Aa	±0.001	0.035Aa	±0.004	0.051Aa	±0.014
Ce	0.053Ab	±0.007	0.023Ac	±0.009	0.464Aa	±0.080	0026Ab	±0.004	0.358Aa	±0.069	0.236Aa	±0.060
Pr	0.090Ab	±0.013	0.031Ac	±0.011	0.730Aa	±0.116	0.039Aa	±0.016	0.310Aa	±0.051	0.355Aa	±0.095
Nd	0.012Ab	±0.002	0.005Ac	±0.002	0.0.096Aa	±0.016	0.004Aa	±0.000	0.051Aa	±0.010	0.045Aa	±0.012
Sm	0.041Ab	±0.006	0.018Ac	±0.008	0.319Aa	±0.051	0.017Aa	±0.000	0.172Aa	±0.034	0.152Aa	±0.038
Eu	0.004Ab	±0.001	*0.30Ab	±0.000	0.048Aa	±0.009	nd	nd	0.024Aa	±0.006	0.023Aa	±0.007
Gd	0.001Ab	±0.000	0.001Ac	±0.001	0.009Aa	±0.001	nd	nd	0.004Aa	±0.002	0.004Aa	±0.001
Tb	0.007Ab	±0.001	0.004Ac	±0.001	0.037Aa	±0.005	0.003Aa	0.000	0.023Aa	±0.006	0.018Aa	±0.004
Dy	0.001Ab	±0.000	0.001Ac	±0.000	0.006Aa	±0.001	nd	nd	0.003Aa	±0.001	0.002Aa	±0.001
Y	0.005Ab	±0.001	0.003Ab	±0.001	0.025Aa	±0.004	0.002Aa	±0.002	0.013Aa	±0.003	0.012Aa	±0.003
Ho	0.001Ab	±0.001	0.001Ac	±0.000	0.006Aa	±0.001	nd	nd	0.002Aa	±0.001	0.002Aa	±0.001
Er	0.003Ab	±0.001	0.001Ac	±0.000	0.012Aa	±0.002	*0.60Aa	±0.000	0.006Aa	±0.002	0.005Aa	±0.001
Tm	0.001Ab	±0.000	*0.40Ab	±0.000	0.003Aa	±0.000	nd	nd	*0.40Ba	±0.000	*0.30Ba	±0.000
Yb	*0.10Ab	±0.000	*0.60Ab	±0.000	0.010Aa	±0.001	nd	nd	0.004Aa	±0.002	0.004Aa	±0.002
Lu	0.001Ab	±0.000	*0.40Ab	±0.000	0.002Aa	±0.001	nd	nd	*0.20Ba	0.000	*0.10Ba	±0.000
Hf	0.006Ab	±0.001	0.003Ab	±0.001	0.031Aa	±0.005	0.001Ab	±0.000	0.010Aa	0.001	0.013Aa	±0.005
Ta	0.008Aa	±0.005	0.003Aa	±0.003	0.002Aa	±0.001	0.004Aa	±0.001	0.04Aa	0.000	0.003Aa	±0.001
Th	0.038Ab	±0.004	0.015Ac	±0.003	0.343Aa	±0.052	0.007Ab	±0.001	0.101Aa	0.015	0.145Aa	±0.031
U	0.009Ab	±0.001	0.003Ac	±0.002	0.068Aa	±0.012	0.001Ab	±0.001	0.020Aa	0.003	0.030Aa	±0.009

Olives (Table 9) showed values between 20-4 ppm for Rb and 3-1 ppm for Sr. Depending on the treatment and geographical origin, trace elements showed high variability. Rb was the only element that highlighted meaningful differences between the two sites, showing higher values for MN than SL. All other elements showed concentrations between 1-0.1 ppb. Concerning trace elements from

MN olives, almost all elements of ZN-DM had higher values, followed by DM and CNT. In SL olives, Sr, Hf, Ta and U revealed no differences between treatments, while all other elements indicated higher values in ZN and ZA treatments than in SF. The stronger contribution of natural zeolite in increasing the concentration of trace elements by a factor 10-100 than traditional treatments is evident also in the olive samples from both sites. The olives results showed values lower than in the literature but generally much lower than leaves. The REE plant uptake is not yet known because only a few studies were performed in natural conditions (Agnan et al., 2014; Brioschi et al., 2013; Punturo et al., 2018); despite that, our data highlighted a trace elements concentration that decreased in the sequence soil>leaf>olive (Brioschi et al., 2013; Pošćić et al., 2020; Turra, 2018; Tyler, 2004).

Table 9. Trace elements composition (ppm) of olive samples (mean ± standard deviation). Values with "*" are expressed in ppb. The values are the result of three replicates. MN and SL are Montiano and San Lazzaro areas. In the MN field, CNT refers to the negative control, DM refers to Dimethoate treatment while ZN-DM refers to natural zeolite + Dimethoate treatment. In SL field, SF refers to the traditional treatment of Spinosad+Spyntor fly, ZA is referred to the NH4+-enriched zeolite and ZN is natural zeolite treatment. "nd" is not detected value. Different uppercase letters indicate significant differences due to the geographical origin while different lowercase letters indicate significant differences between the different treatments within the same experimental site (p<0.05) as a result of ANOVA and Tukey's HSD test.

	MN						**SL**					
REE	**DM**		**CNT**		**ZN-DM**		**SF**		**ZN**		**ZA**	
	mean	**dev std**	**mean**	**dev std**	**mean**	**dev std**	**mean**	**dev std**	**mean**	**dev std**	**mean**	**dev std**
Rb	18.050Aa	±4.247	14.167Aa	±2.690	19.200Aa	±1.567	9.361Bab	±3.469	10.7818Ba	±1.890	4.360Bb	±1.434
Sr	1.953Aa	±0.592	2.008Aa	±0.826	2.046Aa	±0.508	1.230Aa	±0.552	2.247Aa	±0.471	1.764Aa	±0.185
Zr	0.001Ab	±0.001	0.001Ab	±0.001	0.005Aa	±0.001	*0.40Ab	±0.000	0.002Aa	±0.001	0.002Aa	±0.001
Nb	0.009Ab	±0.005	0.003Ab	±0.003	0.048Aa	±0.013	0.004Ab	±0.003	0.031Aab	±0.019	0.023Aa	±0.004
La	0.001Ab	±0.001	nd	nd	0.009Aa	±0.009	*0.40Ab	±0.002	0.002Aa	±0.001	0.002Aab	±0.001
Ce	0.001Ab	±0.001	*0.10Ab	±0.000	0.012Aa	±0.002	*0.95Ab	±0.000	0.006Aa	±0.001	0.005Aa	±0.003
Pr	0.004Ab	±0.003	*0.10Ab	±0.000	0.038Aa	±0.006	nd	nd	0.001Aa	±0.000	0.001Aa	±0.001
Nd	*0.30Ab	±0.000	nd	nd	0.003Aa	±0.001	nd	nd	0.011Aa	±0.005	0.009Aa	±0.003
Sm	0.001Ab	±0.000	*0.20Aab	±0.000	0.011Aa	±0.002	nd	nd	0.006Aa	±0.003	0.005Aa	±0.002
Eu	*0.20Ab	±0.002	*0.10Ab	±0.000	0.002Aa	±0.000	nd	nd	0.001Aa	±0.001	0.001Aab	±0.000
Gd	*0.20Ab	±0.000	*0.20Ab	±0.000	*0.50Aa	±0.000	*0.10Ab	±0.000	*0.50Aa	±0.000	*0.30Aa	±0.000
Tb	*0.40Ab	±0.000	*0.10Ab	±0.000	0.002Aa	±0.000	nd	nd	0.001Aa	±0.001	*0.70Aa	±0.000
Dy	nd	nd	nd	nd	*0.20Aa	±0.000	nd	nd	*0.30Aa	±0.000	*0.10Aa	±0.000
Y	*0.20Ab	±0.000	0.10Ab	±0.000	0.001Aa	±0.000	nd	nd	*0.40Aa	±0.000	*0.50Aa	±0.000
Ho	nd	nd	nd	nd	*0.20Aa	±0.000	nd	nd	0.001Aa	±0.001	*0.10Aa	±0.000
Er	*0.10Ab	±0.000	nd	nd	*0.70Aa	±0.000	nd	nd	0.001Aa	±0.001	*0.30Aa	±0.000
Tm	nd	nd	nd	nd	*0.10Aa	±0.000	nd	nd	*0.20Aa	±0.000	nd	nd
Yb	*0.20Aab	±0.000	nd	nd	0.001Aa	±0.000	*0.10Ab	±0.000	0.002Aa	±0.002	0.40Aab	±0.000
Lu	nd	nd	nd	nd	nd	nd	nd	nd	*0.20Aa	±0.000	*0.10Aa	±0.000
Hf	0.001Ab	±0.000	nd	nd	0.002Aa	±0.000	*0.30Aa	±0.000	0.001Aa	±0.001	0.001Aa	±0.000
Ta	0.003Aa	±0.003	0.002Aa	±0.001	0.002Aa	±0.002	0.001Aa	±0.000	0.002Aa	±0.001	0.002Aa	±0.001

Th	0.001Ab	±0.000	*0.50Ab	±0.000	0.006 Aa	±0.001	nd	nd	0.003Aa	±0.001	0.003Aa	±0.001
U	*0.20Aab	±0.000	*0.30Ab	±0.000	0.002Aa	±0.001	*0.10Aa	±0.000	0.001Aa	±0.000	*0.50Aa	±0.000

Overall, the use of zeolitites as foliar treatment influenced trace elements composition in leaf and olive samples. The application of zeolitite maybe provided these elements to the plants, changing their geochemical fingerprint and allowing to identify the foliar management, as an "induced geochemical traceability" of the final products.

4.3 Discrimination of the geographical area through geochemical characterization and statistical analysis: the comparison of MN and SL fields

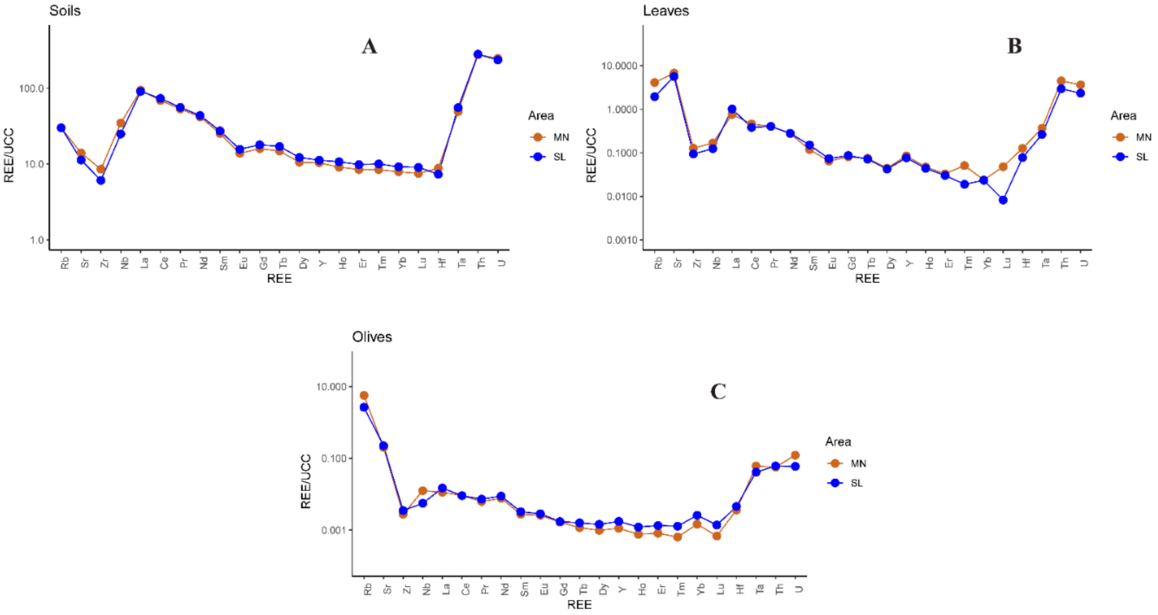

Figure 17. Trace elements of MN (Montiano) and SL (San Lazzaro) sites in (**A**) soils; (**B**) leaves and (**C**) olives. Values are normalized by UCC (Upper Continental Crust).

MN and SL sites (Figure 17A) showed the same trend of trace elements in soils, with generally strong positive spikes of La, Th and U, while Zr always showed a negative anomaly. Although MN and SL are characterized by different textural formation, both sites belong to the "Supersintema Emiliano-Romagnolo" of the "Post-evaporic succession of the Po-Adriatic margin", which means alluvial and deltaic deposits in cyclic successions. By comparing the leaves from the two experimental site it is possible to observe very similar trends (Figure 17B). However, Lu showed a negative anomaly in SL site that was not recorded in MN. The trend recorded from olive samples (Figure 17C) was also very similar for both areas with few differences: i) SL showed a slightly higher trend than MN; ii) MN showed more positive Rb, Nb, Ta and U anomalies than SL.

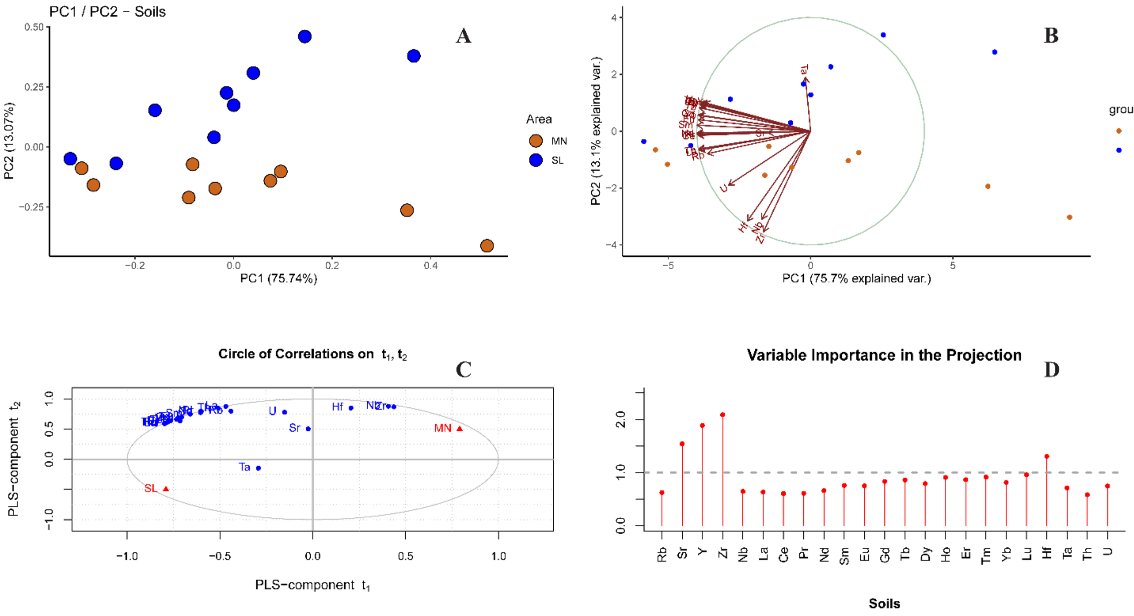

Figure 18. MN (Montiano) and SL (San Lazzaro) soil samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). Observation axes PC1 and PC2: 88.81%.

Comparing the soils from the two sites, PCA total variance showed 88.81% of the observation axes PC1 and PC2: PC1 explained 75.74% and PC2 13.07% of the total variance (Figure 18A). All elements had a negative correlation in the first PC; most elements showed a positive correlation in the second PC, except La, Rb, U, Hf, Nb and Zr which had a negative correlation (Figure 18B). PLS-DA of Figure 18C showed good separation between the groups (MN and SL), in both principal components; Figure 18D highlighted Sr, Y, Zr and Hf elements as a discriminant in the separation of VIP analysis.

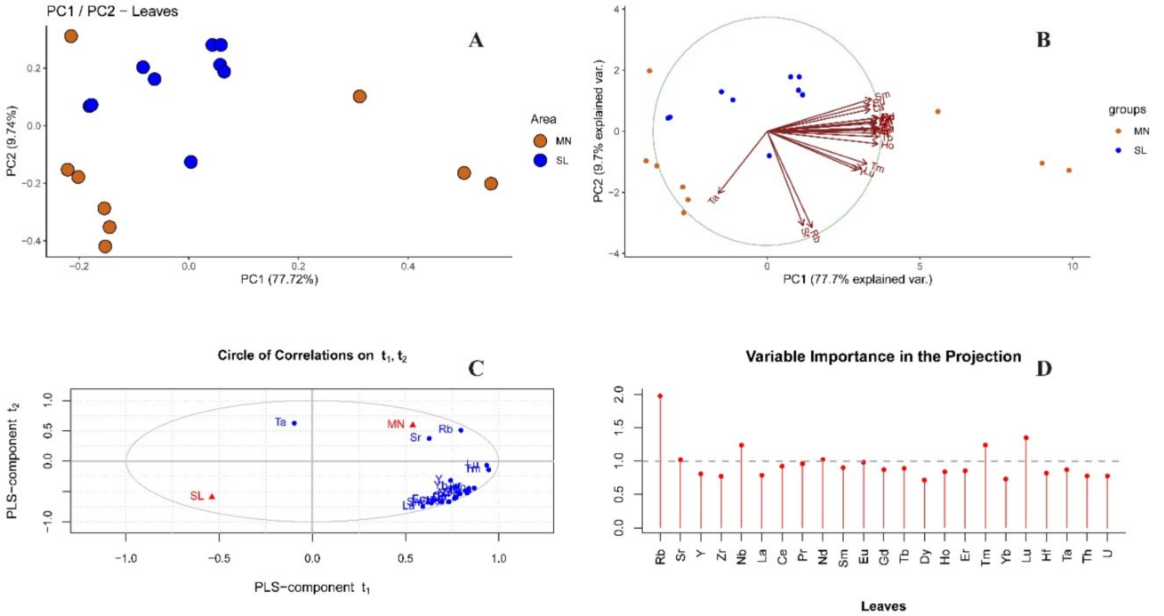

Figure 19. MN (Montiano) and SL (San Lazzaro) leaf samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). Observation axes PC1 and PC2: 87.46%.

In the comparison of leaves from the two areas, the observation axes PC1 and PC2 explained 87.46% of the total variance (Figure 19A), divided into 77.72% of PC1 and 9.74% of PC2. Only Ta had a negative correlation in the first PC; Ta, Sr, Rb, Y, Lu, Tm, Ho and Tb had a negative correlation in the second PC, whereas all other elements showed a positive correlation (Figure 19B). Also, PLS-DA in Figure 19C seemed to confirm the trend of these elements in the separation of the groups (MN and SL); in Figure 19D, VIP analysis indicated the following elements as discriminant: Rb, Sr, Nb, Nd, Tm and Lu.

The model generated by PCA in olive data explained 83.50% of the total variance (66.28% of PC1 and 17.19% of PC2). The first PC showed a positive correlation between trace elements, whereas in PC2 some elements showed a positive correlation (such as Gd, Tb, Dy, Tm) while others a negative correlation (such as Sr, Rb, Nb, U, Ta, Ce, Nd, Sm,U) (Figure 20B). PLS-DA of the comparison of the two areas (Figure 20C) showed a good split-up between MN and SL samples, although the difference was not so clear as for soils and leaves. La, Nd, Sm, Eu, Dy, Ho, Yb Lu, Hf and Th were detected as the most relevant influencers for the discrimination in the VIP analysis (Figure 20D).

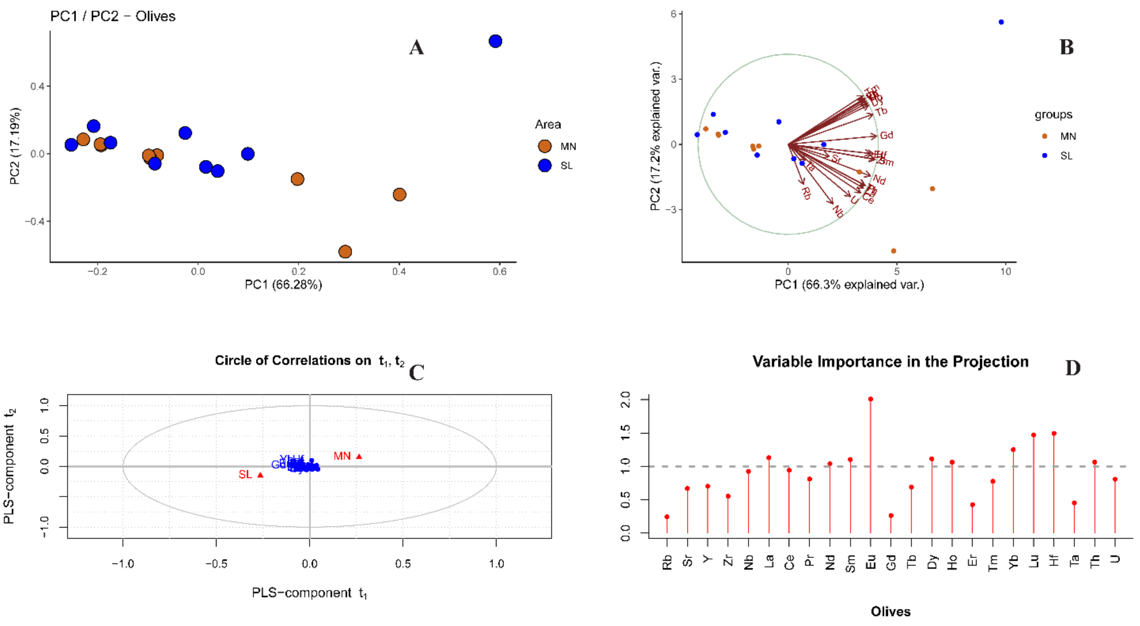

Figure 20. MN (Montiano) and SL (San Lazzaro) olive samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). Observation axes PC1 and PC2: 83.50%.

Overall, PCA and PLS-DA showed good separation between MN and SL sites with a better separation for soils and leaves than for olives. From the statistical analyses, the only elements useful for the correlation of soil, leaf and olive samples for the discrimination and the identification of the geographical origin were Lu and Hf, which were identified as the best discriminator elements for the traceability of this study. Lu and Hf elements belong to, respectively, the Heavy REE (HREE) and High Field Strength Elements (HFSE) groups and their content may depend on the geology of the areas studied: Lu is usually found in trace amounts in rare-earth element-loving minerals (garnet and phosphates), whereas Hf can be stored in trace amount in zirconium-rich minerals. Thanks to their properties, REE are also important as tracers for geochemical processes (Henderson, 1984; Lipin B. and McKay G., 1989). As reported in the geological description, MN and SL were characterized by lithological variability, having clay and sandy-loam/clay-loam fractions, respectively. The various amounts of terrigenous fractions and the presence of different minerals with these trace elements in rocks, linked to the partial variability in lithology of the two areas, can explain the presence of Hf and Lu also in soils and plants. Moreover, the microclimate of each site, especially the precipitations, may influence the weathering processes and hence the availability of certain elements. In 2019, the recorded annual precipitations in the Bologna and Forlì-Cesena provinces were respectively around +100mm and -100mm with respect to the average (years 1961-1990) for the two areas. Overall, SL (Bologna province) showed a positive anomaly (around 800mm of precipitations with +23 rainy days) whereas MN (Forlì-Cesena province) had a negative trend (700mm of precipitations and -20 rainy days) compared to the 1961-1990 climatic data. The hydroclimatic balance (difference between precipitation and evapotranspiration) also highlighted differences between the two sites: Forlì-Cesena showed a negative anomaly of hydroclimatic balance stronger than Bologna (-200mm and -20mm, respectively), which means that in the Forlì-Cesena province the evapotranspiration was larger than in Bologna area (Pavan and Marletto, 2020). This parameter contributed to the microclimatic variability of the two sites and consequently to the geochemical composition of final products.

Table 10. The Bioaccumulation Coefficient (BA) and the Traslocation Coefficient (TC). MN and SL are Montiano and San Lazzaro areas.

	BA: C_{leaf}/C_{soil}		TC: $C_{solid\ residue}/C_{leaf}$	
	MN	**SL**	**MN**	**SL**
Rb	0.137	0.065	1.380	1.355
Sr	0.483	0.498	0.030	0.040
Zr	0.015	0.015	0.022	0.037
Nb	0.005	0.005	0.075	0.045
La	0.008	0.011	0.015	0.015
Ce	0.007	0.005	0.020	0.024
Pr	0.008	0.007	0.016	0.018
Nd	0.007	0.006	0.028	0.032
Sm	0.005	0.006	0.023	0.021
Eu	0.005	0.005	0.040	0.039
Gd	0.005	0.005	0.021	0.020
Tb	0.005	0.004	0.016	0.022
Dy	0.004	0.003	0.022	0.034
Y	0.008	0.007	0.013	0.023
Ho	0.005	0.004	0.016	0.028
Er	0.004	0.003	0.025	0.044
Tm	0.006	0.002	0.013	0.068
Yb	0.003	0.003	0.061	0.111
Lu	0.006	0.001	0.014	0.168
Hf	0.014	0.011	0.029	0.059
Ta	0.007	0.005	0.168	0.156
Th	0.016	0.010	0.013	0.021
U	0.015	0.010	0.033	0.026

The BA coefficient had values between 1-0.1 for Rb from MN and Sr from both sites; 0.1-0.01 for Rb and U from MN, La from SL and Zr, Hf and Th from both sites; <0.01 for all other elements. The TC coefficient showed values between 1-0.1 for Rb and Ta from both fields, but Yb and Lu from SL; 0.1-0.01 for all other elements.

According to BA results, Rb and Sr were the elements preferentially accumulated from soils to leaves in MN site, whereas Sr was preferentially accumulated from soils to leaves in SL field. Sr is an element mainly present in trace concentration in rocks and soils, but it also occurs in plants at a lower concentration, depending on the species. Sr in plants is assumed to be absorbed from the soil mainly during germination (Medini et al., 2015) and plants can use different sources of Sr from soil: Sr in organic or inorganic complexes (Capo et al., 1998), Sr in free solution (Semhi et al., 2012), or Sr from the underlying groundwater. On the other hand, no explanation was found for the high Rb content in MN and SL sites, although the TC had the highest correlation for both sites (Figure 21). From the analyses described above, Rb in leaf and olive showed a linear correlation and both variables were normally distributed (Shapiro test p>0.05).

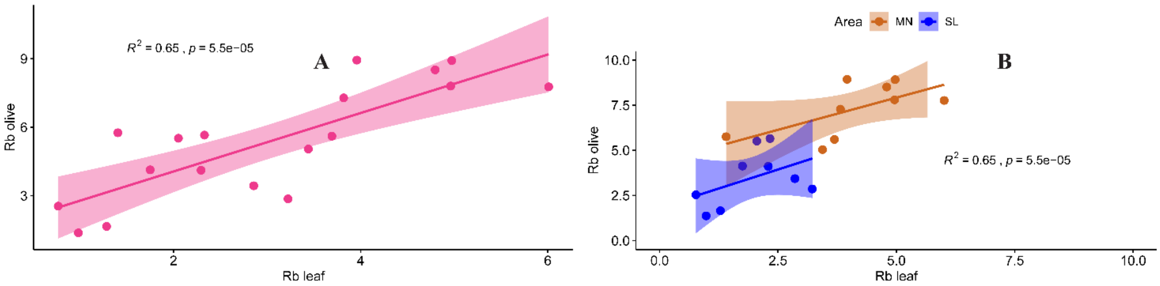

Figure 21. Correlation between Rb content in leaf and olive samples for (**A**) both areas (R^2=0.65) and (**B**) MN (Montiano) and SL (San Lazzaro) sites. Confidence level is 95%.

Rb showed the highest correlation in leaf and olive samples. The total Pearson correlation showed an R index of 0.81, which means an R^2=0.65 ($p<0.001$) (Figure 21A): apparently, Rb could be accumulated more in leaves and olives than in the soil from where plants have grown, as shown in higher values of this element in TC coefficient than in BA index (Table 10). Figure 20B highlighted that MN site contributed more than SL field to this correlation: MN presented an R index of 0.63, which means R^2=0.34, while SL had an R index of 0.46, which correspond to R^2=0.21, but their p-values were not meaningful. On the other hand, the BA index of Sr had a higher concentration than TC coefficient (Table 10), which means that Sr tends to accumulate more in soil and plant than in the final product.

Both Rb and Sr contents, strongly depend on the geochemical composition of the soil of origin (Giaccio and Vicentini, 2008). Although the samples came from a limited geographical area, they showed a meaningful difference between the two sites of the study, contrary to the work of Nasr et al., 2021 that found a less significant difference in olive samples from North Spain and South France countries due to the limited and restricted geographical area of his study.

Overall, Lu and Hf were the best discrimination elements to identify the geographical fingerprint of samples from MN and SL sites. However, as already explained, also Rb and Sr had a meaningful role in the plant uptake.

4.4 Effects of different foliar treatments on the geochemical fingerprints of leaves and olives in MN and SL sites

MN site

The elements pattern recorded in MN area was significantly different between the various treatments tested (Figure 22). Concerning leaf samples (Figure 22A), Rb, Sr, La, Th and U seemed to have strong positive anomalies, whereas Sm and Yb had meaningful negative anomalies in CNT and DM treatments respectively. By looking into olives data (Figure 22B), pronounced Rb and Sr positive anomalies can be observed as well as Hf, Ta and U slight positive anomalies. A strong negative anomaly in Tb and a slight negative anomaly in Nb were also evident for the CNT treatment.

On average, it is clear that higher values were always recorded in the treatment in which zeolite minerals were employed (ZN-DM), followed by DM treatment, while the lowest values were recorded in the CNT. This trend has already been shown in Table 7 and Table 8 and it can due to the contribution of zeolite minerals and volcani glass that provided trace elements to the plant, allowing a clear distinction and a good geochemical separation between foliar treatments.

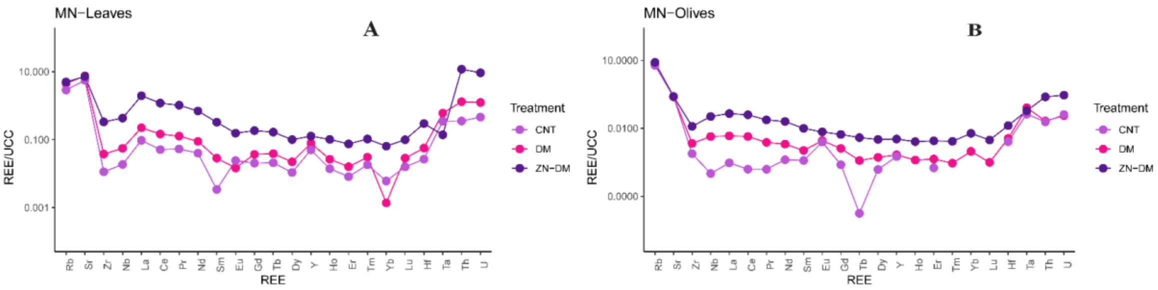

Figure 22. Trace element in MN (Montiano) site for (**A**) leaves and (**B**) olives. In the MN field, CNT is referred to as negative control, DM means Dimethoate and ZN-DM is natural zeolite + Dimethoate. Values are normalized by UCC (Upper Continental Crust).

The PCA of leaves from MN generated 95.64% of the total variance, divided as follows: 87.98% of PC1 and 7.66% of PC2 (Figure 23A). The PCA loading plot of Figure 23B highlighted a positive first PC for all elements, except Ta; PC2 showed Ta, Sr, Rb and Y negative correlation, whereas all other elements had a positive correlation. In PLS-DA of MN (Figure 23C), treatments seemed to have a good separation, especially ZN-DM against CNT and DM, and therefore was able to discriminate samples according to leaf practices. VIP analysis (Figure 23D) confirmed that Rb, Sr, Sm, Dy and Ta can be the elements used to identify groups with different treatments.

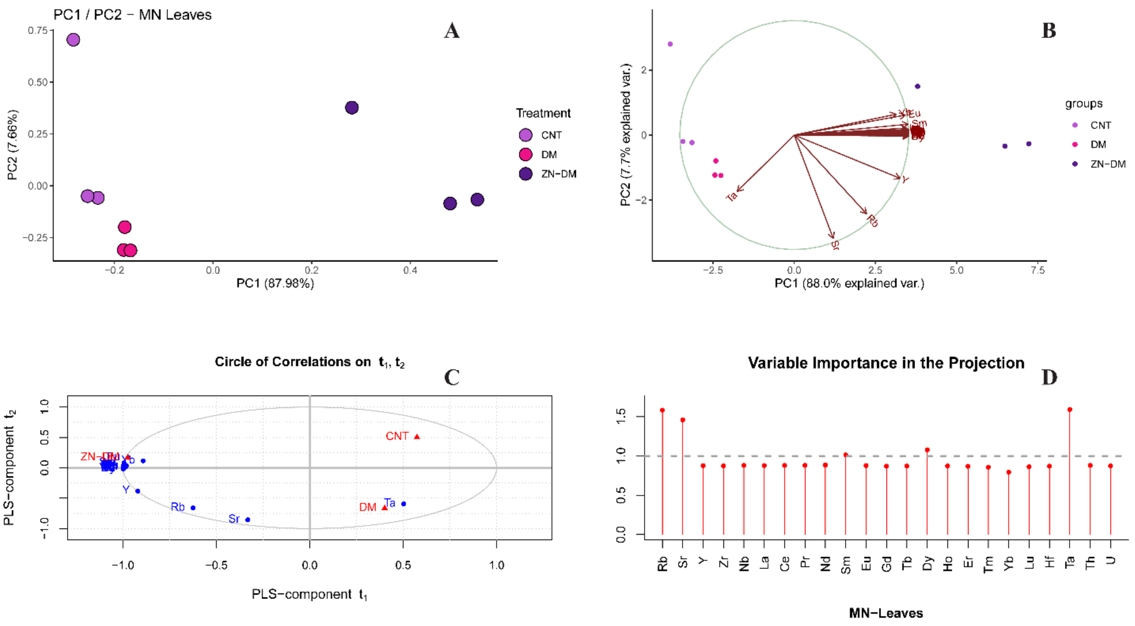

Figure 23. MN (Montiano site) leaf samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). CNT is referred to as negative control, DM means Dimethoate and ZN-DM is natural zeolite + Dimethoate. Observation axes PC1 and PC2: 95.64%.

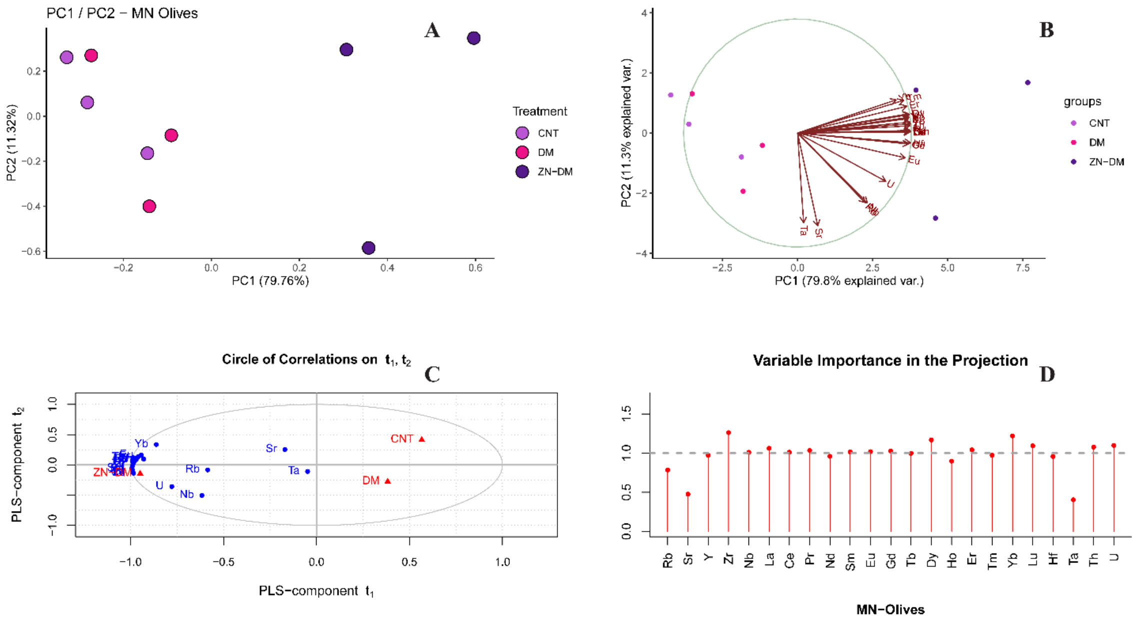

Figure 24. MN (Montiano site) olive samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). In the MN field, CNT is referred to as negative control, DM means Dimethoate and ZN-DM is natural zeolite + Dimethoate. Observation axes PC1 and PC2: 91.08%.

The PCA of olives from MN generated 91.08% of the total variance, divided into 79.96% of PC1 and 11.32% of PC2. The PCA loading plot (Figure 24B) showed a positive first PC for all elements; also the PC2 indicated a positive correlation for all trace elements, except for Ta, Sr, Rb, U, Eu and Nb which had a negative correlation. In the MN area, PLS-DA highlighted a meaningful separation between ZN-DM and the other two treatments (Figure 24C). VIP analysis (Figure 24D) showed the following elements as identifiers: Zr, Nb, La, Ce, Pr, Sm, Eu, Gd, Tb, Dy, Er, Yb, Lu, Th and U.

Overall, PCA and PLS-DA allowed to well discriminate the ZN-DM treatment and VIP analyses identified Sm and Dy as the most relevant elements for the correlation of both leaves and olives for the discrimination treatments in MN. Esser et al. (1991) have correlated the presence of Sm, as well as La, Ce, Nd, Eu and Tb, to the originating Fe-rich chlorite leached from E- and B-horizons in soils from the Indiana Dunes (USA). The uptake of Dy element could be significantly related to the organic matter-bound concentrations (Tyler, 2004; Xinde et al., 2000).

Trace elements are essential micronutrients that are required for the healthy growth of plants (Jagadeesha et al., 2018). Although plants accumulate trace elements mainly from root uptake, it is not unusual for the process to take place through foliar absorption. This could confirm the hypothesis that foliar treatments, especially the use of zeolitite, may provide some trace elements to the plant (leaf and olive) and modify the geochemical fingerprint of the final product.

SL site

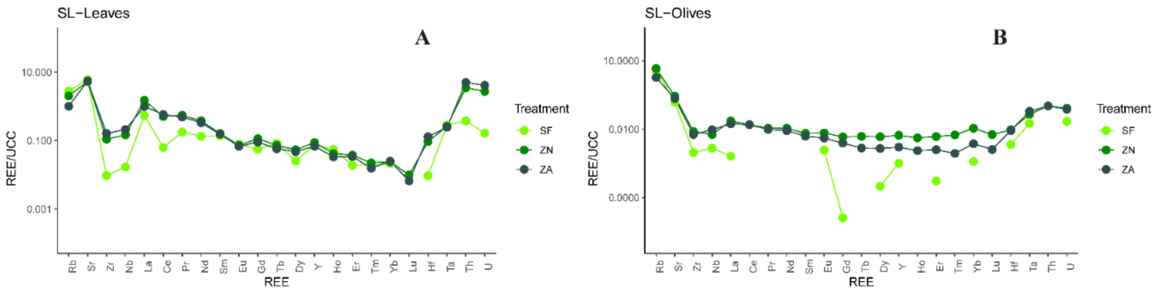

Figure 25. Trace element of SL (San Lazzaro site) of (**A**) leaves and (**B**) olives. In the SL field, SF is the traditional treatment of Spinosad+Spyntor fly, ZA is referred to the NH_4^+-enriched zeolite and ZN is natural zeolite. Values are normalized by UCC (Upper Continental Crust).

As for MN area, also in SL significant differences ($p<0.05$) between the foliar treatment were found (Figure 25). The pattern observed in SL was similar to that of MN but the concentrations recorded in the olives were an order of magnitude greater.

Concerning leaf samples, almost all elements showed a higher trend for ZN and ZA treatments than the conventional SF. Rb, Sr, La, Th and U had significant positive anomalies, on the contrary, Lu showed very low values and the elements among Ce and Yb presented a trend with decreasing values (Figure 25A). On the other hand, the SL olives (Figure 25B) highlighted a Gd strong negative anomaly in SF treatment; in SF treatment, there also is an Nb opposite trend to the other treatments. As occurred in MN site, the highest values were recorded in ZN and ZA olives, where zeolite minerals were employed and probably have contributed to the geochemical fingerprint.

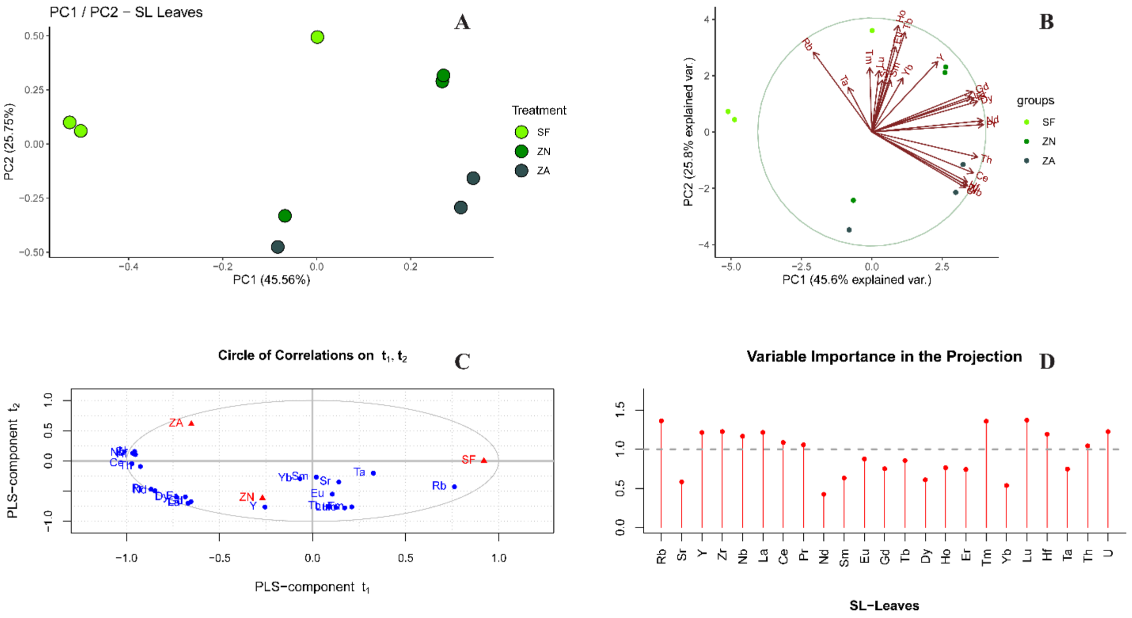

Figure 26. SL (San Lazzaro field) leaf samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). SF is the traditional treatment of Spinosad+Spyntor fly, ZA is referred to as the NH_4^+-enriched zeolite and ZN is natural zeolite. Observation axes PC1 and PC2: 71.31%.

PCA of SL leaves indicated 46.56% of PC1 and 25.75% of PC2, with 71.31% of the total variance (Figure 26A). PCA loading plot (Figure 26B) showed that elements had a positive correlation in PC1 unless Rb, Ta and Tm; meanwhile elements were split into positive (Rb, Ta, Tm, Lu, Ho, Tb, Sm, Yb, Nd, Dy, La, Gd, Y) and negative (Ce, Th, Pr, Hf) correlations in PC2. The PLS-DA (Figure 26C) highlighted a meaningful separation between SF and other treatments with zeolite, while a lower separation in the secondary component allows to distinguish ZA from ZN. VIP analysis (Figure 26D) pointed out Rb, Y, Zr, Nb, La, Ce, Pr, Tm, Lu, Hf, Th and U as discriminant elements.

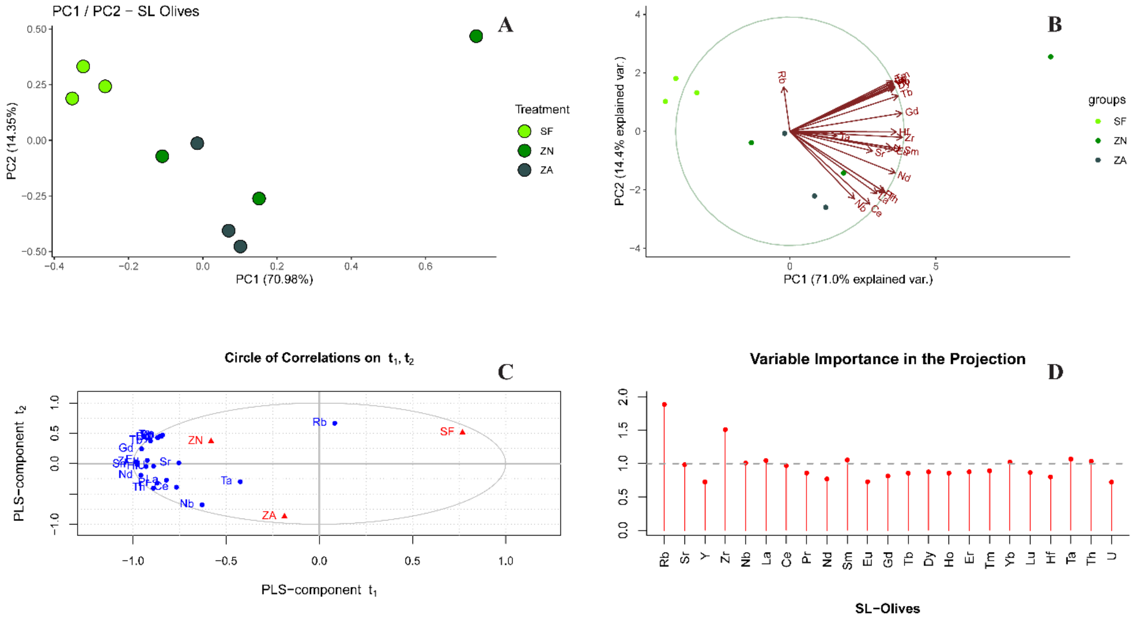

Figure 27. SL (San Lazzaro site) olive samples: (**A**) PCA score plot and (**B**) PCA loading plot; (**C**) PLS-DA score plot and (**D**) Variable Importance in the Projection (VIP). SF is the traditional treatment of Spinosad+Spyntor fly, ZA is referred to as the NH_4^+-enriched zeolite and ZN is natural zeolite. Observation axes PC1 and PC2: 85.33%.

Olives from SL indicated 85.33% of the total variance, divided into 70.98% of PC1 and 14.35% of PC2. All trace elements had a positive correlation in PC1, except Rb; the secondary PC was divided equally into positive (such as Rb, Tb, Gd, Hf) and negative (Zr, La, Sr, Nd, Ta, Ce, Nb) correlations. Also PLS-DA score plot of SL (Figure 27C) pointed out an important separation among treatments and Rb seemed to be the best element to distinguish the groups; a good separation in the secondary component (PC2) also occurred between ZN and ZA. Furthermore, VIP (Figure 27D) proved that Rb, Zr, Nb, La, Sm, Yb, Ta and Th are relevant for the discrimination among treatments.

Overall, SL area gave a significant separation between treatments with zeolitite (ZN and ZA) and the traditional SF, probably thanks to the influence of zeolite on the geochemical fingerprint of leaf and olive, as already explained for MN site. However, in PCA it was not possible to distinguish the natural zeolite (ZN) and the enriched one (ZA) and only the PLS-DA of SL pointed out a slight separation between ZN and ZA for both leaves and olives. Given that VIP analyses highlighted Rb, Zr, La and Th as the only elements in both leaf and olive samples, they were used as discriminator elements to correlate leaves and olives from the SL area.

4.5 The olive oil traceability

C isotopic signature for all FAs (Figure 28) showed partial discrimination of olive oils due to the different geographical origins. All values were included between -20 and -40‰ of $\delta^{13}C$, although they revealed a meaningful variability. Brisighella (BG) highlighted the highest values, while Ancona (AN), Bologna (BO) and Pescara (PE) had the more negative $\delta^{13}C$. Figure 29 shows in detail the most abundant FAs and the difference that occurred among localities. Apparently, BG showed more positive values than other sites for each FA, normally followed by SL and PE. Generally, other sites highlighted a high variability according to the FA: PE showed high results in C16:0 (Figure 29A) FA, while SL in C16:1, C:18, C18:1w9c (Figures 29B, C, and D). In general, AN, BO and MN sites showed quite low values compared to other sites. A possible reason can be found in i) the different foliar treatments applied and climate conditions, which already influenced the geochemistry of olives or ii) the production methods of olive oils, which can be by hot or cold extraction. Microclimate and foliar treatments, linked to the lithology and soil type, can influence the leaching and plant uptake of some elements. Moreover, the products used as foliar treatments probably had a C fingerprint that we can find in olives and consequently in olive oils, despite the zeolite minerals do not present any carbon in their mineralogical composition. Cold extraction generally ensures a higher oil quality, helping to increase its organoleptic qualities and to preserve healthy olives. By using cold extraction methods, extra virgin olive oil producers favour quality over quantity.

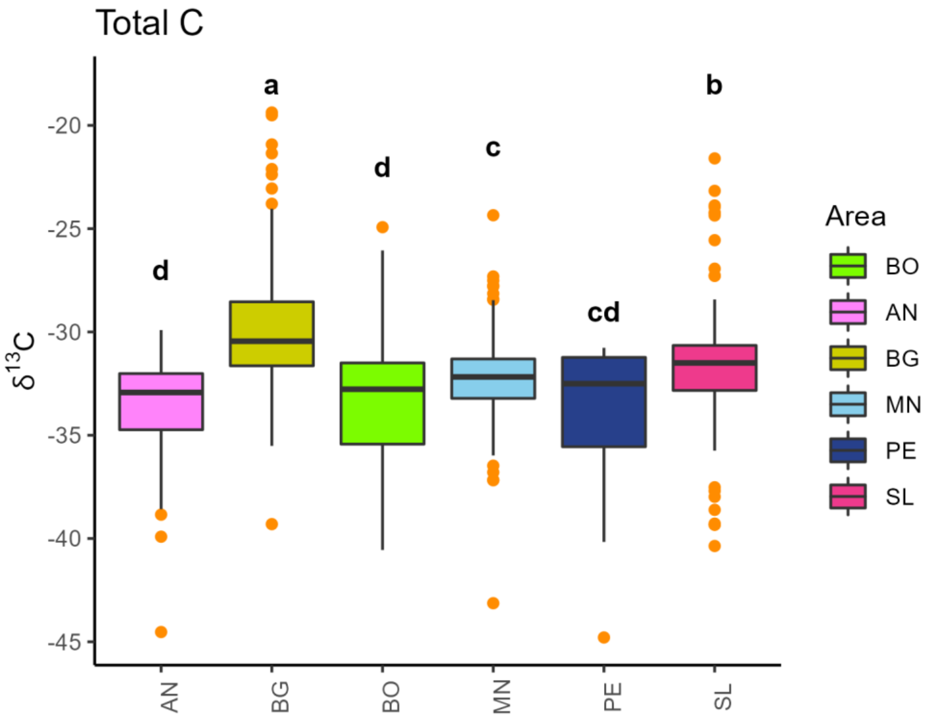

Figure 28. Box-plot of $\delta^{13}C$ for olive oil samples. Samples are divided by geographical origin: Ancona (AN), Brisighella (BG), Bologna (BO), Montiano (MN), Pescara (PE) and San Lazzaro di Savena (SL). Different letters represent significant differences ($p<0.05$) as a result of ANOVA and Tukey (HSD) tests.

The model generated by PCA in olive oil data (Figure 30A) explained 69.61% of the total variance, with 50.51% of PC1 and 19.10% of PC2. PLS-DA of the comparison of all areas (Figure 30B) showed a meaningful separation of BG and SL from other sites among PLS-component t_1, while MN highlighted less differentiation compared to all other experimental fields in the PLS-component t_2. Among the most abundant FAs in olive oils, C18:0 was the best discriminator in VIP analysis, followed by C18:3w6,9c and C16:1 (Figure 30C). Also, C16:0, C18:1w9c and C18:2w6,9c fatty acids were relevant influencers for the analysis.

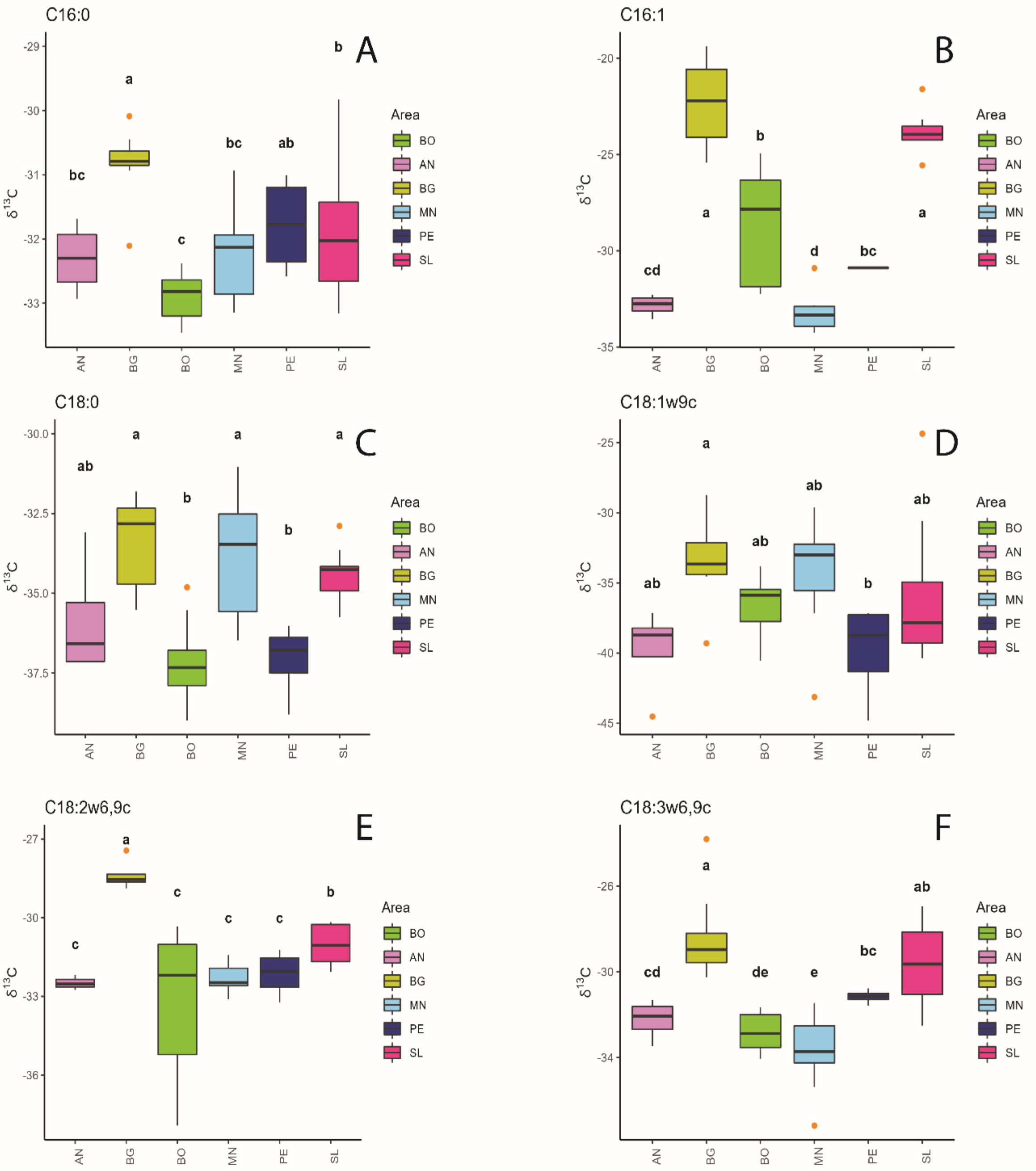

Figure 29. Boxplots of δ^{13}C in olive oils for **(A)** C16:0, **(B)** C16:1, **(C)** C18:0, **(D)** C18:1w9c, **(E)** C18:2w6,9c and **(F)** C18:3w6,9c fatty acids. Samples are divided by geographical origin: Ancona (AN), Brisighella (BG),

Bologna (BO), Montiano (MN), Pescara (PE) and San Lazzaro di Savena (SL). Different letters represent significant differences ($p<0.05$) as a result of ANOVA and Tukey (HSD) tests.

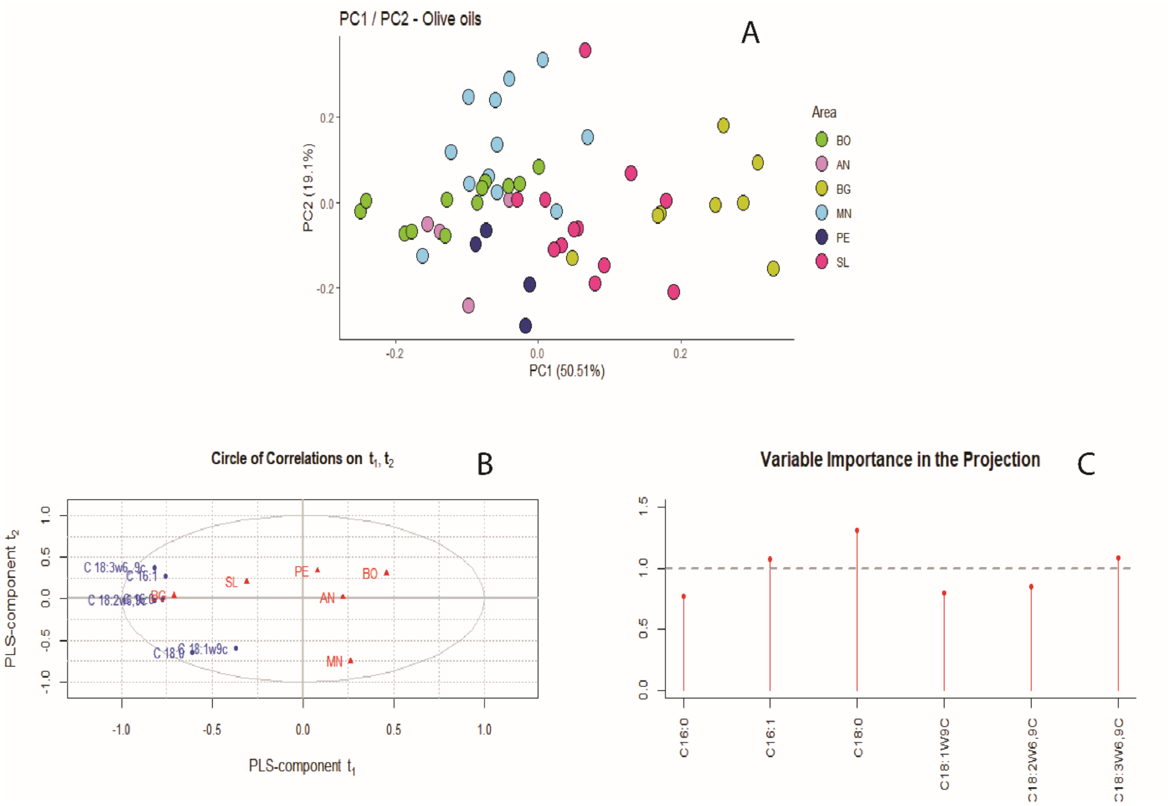

Figure 30. $\delta^{13}C$ for total C fatty acids: (**A**) PCA score plot; (**B**) PLS-DA score plot and (**C**) Variable Importance in the Projection (VIP). Samples are divided by geographical origin: Ancona (AN), Brisighella (BG), National Research Council in Bologna (BO), Montiano (MN), Pescara (PE) and San Lazzaro di Savena (SL). Observation axes PC1 and PC2: 69.61%.

It is well known that FAs composition strongly depends on morphological and biogenetic characterization, and the genotype represents the most important olive oils discriminator (D'Imperio et al., 2007a). In other words, the cultivar affects the similarities/differences among FAs composition accordingly to the genotype. In this study, the cultivars' effects did not take into account, although this parameter had a strong effect on the identification of FAs composition. Indeed, oils from AN and PE came from a mix of olives cultivars, while monovarietal olives were used to produce oils from BO (*Cv Correggiolo*), BG (*Cv Nostrana di Brisighella*), MN (*Cv Correggiolo*) and SL (*Cv Correggiolo*). The specific cultivar can be considered conservative in different years or undergoes significant modifications. Boggia et al. (2002) for example linked the olive fly infestation with climatic conditions, given that a "good" climate, such as high summer temperatures and poor autumn rainfalls, limits the infestation and improves olive oil quality. Baccouri et al., (2022) and Lechhab et al., (2022) also discussed how temperature, humidity and conditions of oil production can affect the FAs composition. This suggests that, although the effect of the cultivar is predominant in the olive oil classification based on the fatty acid composition, a minor geographic effect also exists (D'Imperio et al., 2007a). The FAs composition varies depending on where the crop is grown, mainly due to environmental factors (Al-Bachir and Sahloul, 2017; Bensmira et al., 2007). Fatty acids with 18 C atoms, such as oleic and linoleic fatty acids, play a fundamental role not only in the cultivar characterization but also in the pedoclimatic characterization, accordingly to the previous work of D'Imperio et al., (2007a). Some other variables can affect the FAs composition in olive oils, such as the irrigation regime and storage time of olive oils (Al-Bachir and Sahloul, 2017; D'Imperio et al., 2007b). However, in this study, no information has been provided about these parameters and we have assumed that each company prefers to rely on its usual practices and undergoes its own management regime. In conclusion, the difference in FAs composition among olive oils from different areas could be derived from pedoclimatic conditions, variety and climatic conditions that influence drupe ripening and composition (Di Vaio et al., 2021).

In addition to geographical and climatic conditions or further environmental aspects, many other factors, such as agricultural practices (ripeness index, storage conditions, processing), can influence the chemical composition of olive oils (Borges et al., 2017).

Figure 31 shows the results of olive oils PCA to verify the influence of foliar treatments on the FAs composition. The best results of olive oil differentiation were highlighted in the SL site (Figure 31B), showing a total variance of 89.35% (72.63% of PC1 and 16.72% of PC2). MN follows with 71.98% of the total variance, divided as: 51.03% of PC1 and 20.95% of PC2 (Figure 31A). BO generated 71.84% of the total variance, divided by 49.20% of PC1 and 22.34% of PC2 (Figure 31C).

Few works were performed to verify which factors mainly affect olive oil composition, including fertilization (Fernández-Escobar et al., 2006), irrigation regime (Patumi et al., 2015) and agronomic practices in general (Lavee and Wodner, 2015). Although many evidences linking irrigation and FAs composition have been reported (Ayton et al., 2007; Lavee and Wodner, 2015; Patumi et al., 2002), no studies were able to certainly combine FAs and agricultural practices (Mechri et al., 2009). Some research groups also compared organic and conventional practices to discriminate olive oils, but no consistent overall trends were observed (Lanza and Ninfali, 2020; Rosati et al., 2014; Samman et al., 2008). Only De Wit et al. (2016) reported that agricultural practices and growing conditions of the plants could have contributed to the large variation and instability of C18:1c9 observed for some cultivars across seasons. However, the previous works aimed at linking agricultural practices and FAs quantitatively more than qualitatively, unlike this work. Since there are no studies about the effects of foliar treatments on FAs composition in olive oils, it is difficult to explain the reason that allowed the discrimination of different foliar management.

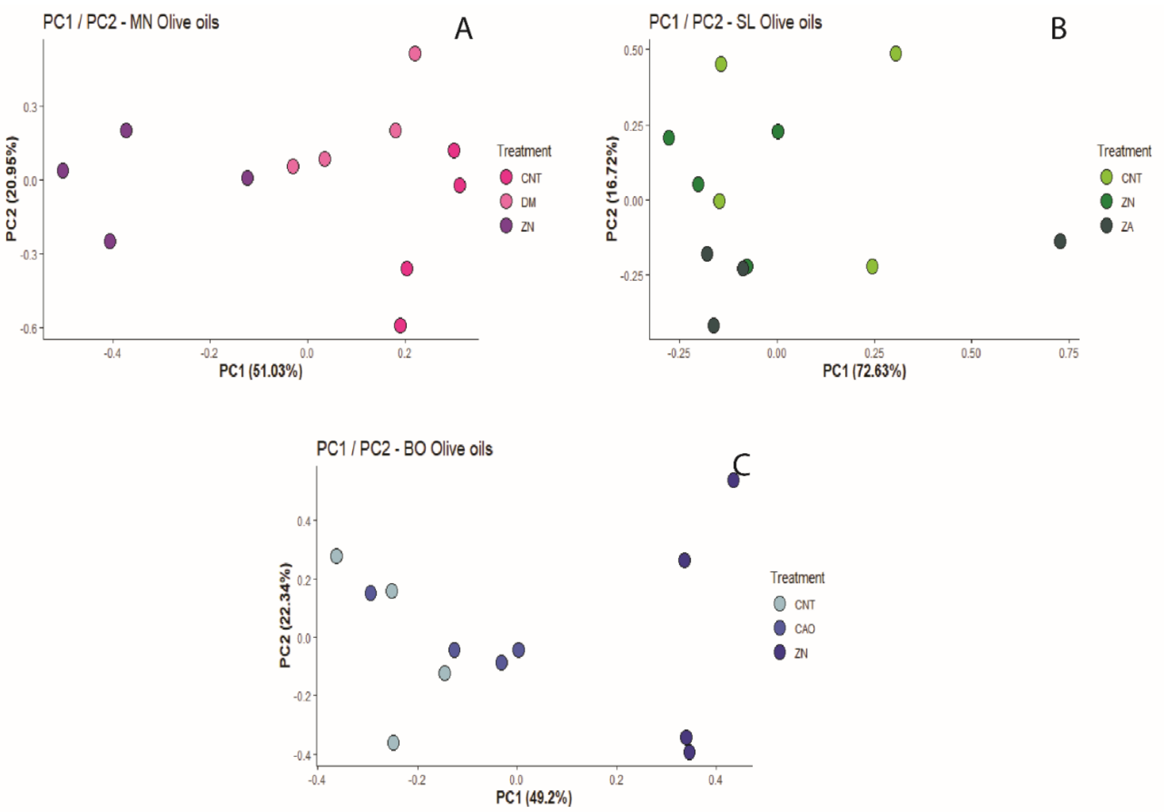

Figure 31. PCA score plot of δ^{13}C for total C fatty acids from (**A**) Montiano (MN); (**B**) San Lazzaro (SL) and (**C**) Bologna (BO). CNT is referred to as negative control, DM means Dimethoate and ZN-DM is natural zeolite + Dimethoate, SF is referred to Spyntorfly + Spinosad, ZN means natural zeolitite, ZA is referred to enriched zeolitite and CAO means Kaolin. Observation axes PC1 and PC2: 71.98% in MN, 89.35% in SL and 71.84 % in BO.

Zeolitite as foliar treatment probably caused a direct or indirect effect on olive oil (or even a combination of both). Chemical and natural products used as foliar treatments probably provided a direct input of C which can influence the signature of leaf and olive, and, consequently olive oil.

On the other hand, it is well known that zeolitite influences plant physiological activities including photosynthesis, with a consequent variation on δ^{13}C (De Smedt et al., 2017; Morrone et al., 2021; Zheng et al., 2018), because during photosynthesis C3-plants discriminate against ^{13}C, causing a fractionation of C that can be appreciated in oils (O'Leary, 1988).

5. Conclusions of olive growing experiment

Thanks to a 3-year experiment conducted in three sites within the Emilia-Romagna region, the efficiency of zeolite minerals in reducing the fertilizer N input up to 50% in olive growing was demonstrated.

N dynamics and all the observed variables were influenced by the fertilizer management (type, amount and timing of application), time, soil texture and irrigation.

However, no differences were observed owing to the different treatments (ZEO and CNT), neither in the detail of each experimental site nor from the general point of view, although in ZEO the fertilization had been reduced by 50%.

The vegetative measurements highlighted a greater development of the olive aerial parts in ZEO treatments compared to CNT. The vegetative measurements conducted in the first year indicate that the plants treated with zeolitite, despite the 50% reduction in fertilizers, have developed similarly to the CNT. In the two rainfed orchards (SL and BN), the ZEO-treated olive plants were characterized by a greater canopy development. Further studies are underway to evaluate the effects, in the long term, of the *una tantum* zeolitite addition as well as the influence on fruit development and the chemical and sensorial quality of the oils.

In conclusion, the use of this specific Italian chabazite zeolitite in olive growing can allow a significant reduction in fertilizer N input, reducing the N losses and improving the plant's physiological status, with meaningful benefits under agronomic, environmental, economic and health aspects. It is indeed very important to specify that the effects of these minerals in the soil are long-lasting due to their long-term structural stability at an ambient temperature and pressure. Moreover, the reduction in the application of N fertilizers can be performed repeatedly over the years, with significant economic and environmental benefits which last forever.

Regarding the geographical identification of olives from SL and MN, the different lithologies played a key role. Hf and Lu appeared to be the best elements for discriminating soils, leaves and olives from the two sites. In this context, the amount of terrigenous fractions and the composition of heavy minerals rich in these two elements (HREE and HFSE), as well as the microclimatic differences of the 2019 agronomic year, provided Hf and Lu also in plants. Moreover, from BA and TC coefficients, Rb and Sr were found to play a meaningful role in plant uptake, depending on the geochemical composition of the soil. Also, C signature in FAs allowed to well discriminate olive oils from different geographical origin, given a large contribute to the Agri-Food traceability thanks to the isotopic composition.

About the discrimination due to the agricultural practices, the trace elements distribution in leaves and olives was higher in zeolite treatments than those without for both MN and SL areas, and the statistical analyses allowed to discriminate the different treatments at each site. FAs of olive oils from MN, SL and BO allowed to separate the treatments according to the C signature, probably due to a direct and/or undirect effect of the foliar treatment on the plant.

Thereby, the use of different foliar treatments, mainly the chemical products such as Dimethoate and Spynosad+Spyntorfly and geological materials such as natural zeolitites, led to a meaningful alteration of the geochemical and isotopic fingerprint in leaves and fruits. Our study led to two important outcomes: i) the importance of agronomic practices at the farm scale in traceability studies given that they influence the geochemical fingerprint of the final products and ii) the possibility of eventually modifying the geochemical fingerprint of a product by using a specific foliar treatment to "personalize" the chemical composition for traceability aims. Future studies are therefore necessary to test more geomaterials (such as kaoline, different types of zeolites etc.) and crops.

6. Results and Discussions of grapevine experiment

6.1. The effects of zeolitite in soil and the water stress conditions

Figure 32A showed a slight acidification tendency among time (from the Pre-Fert sampling to the Harvest) with pH values decreasing from ~8 to 7.5. The acidification of soil is usually a consequence of ion leaching (Mg, K and Ca), due to i) human activities, such as excessive fertilization, ii) intense precipitation or irrigation and iii) nutrient uptake from the plant. The zeolitite application can also cause an increase of soil pH (Mahesh et al., 2018; Polat, E., Karaca, M., Demir, H. and A.N., 2004) and indeed, in the soil treated with zeolitite, a higher pH was recorded (both with and without water stress). Soil Organic Matter (SOM) was in the range 2-7% (Figure 32B) and did not change significantly over the experimentation. SOM and water content influence many microbial processes, including the N mineralization/immobilization which promotes the production or immobilization of NH_4^+ and/or NO_3^-. The combined interactions of SOM and pH can also affect the soil CEC and the NH_4^+ production and dynamics (Eldor, 2007).

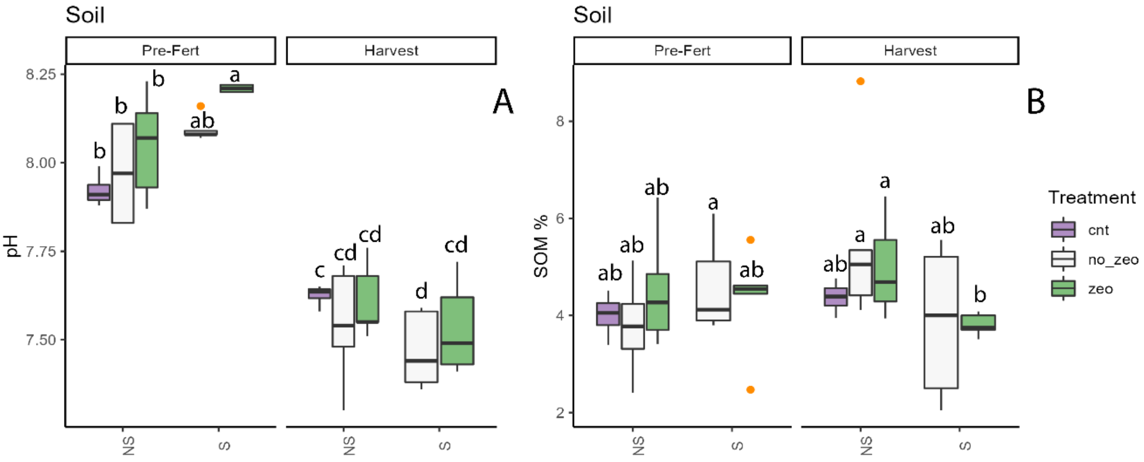

Figure 32. Boxplot of pH (**A**) and Soil Organic Matter (SOM%) (**B**) in soils. NS is No-Stress conditions; S means Water Stress. "Pre-Fert" is the first sampling at Pre-Fertilization time; "Harvest" is the second sampling at Harvest. Different letters represent significant differences ($p<0.05$) as a result of ANOVA and Tukey (HSD) tests.

Total Carbon (TC) and Total Nitrogen (TN) in leaves clearly reflected the effect of water stress (Figure 33). The S treatment highlighted meaningful differences in TC compared with other treatments, with significantly lower values. The lower TC value in S treatment could be due to a deficit in plant growth under water stress conditions. The irrigation and water content in soil have a major effect on photosynthesis and, consequently on plant growth (in terms of foliar area) (Kliewer and Dokoozlian, 2005). A lower plant growth (reduced photosynthesis) implies a lower C acquisition (Vrignon-Brenas et al., 2019). Moreover, treatments with zeolitite showed a higher TC trend, although not statistically significant, than the non-amended soils in the last two samplings. This trend was recorded also in the TN (Figure 33B), given that N status produces effects on grapevine carbon (C) growth and storage, as studied at the annual and pluriannual scales by Vrignon-Brenas et al., 2022.

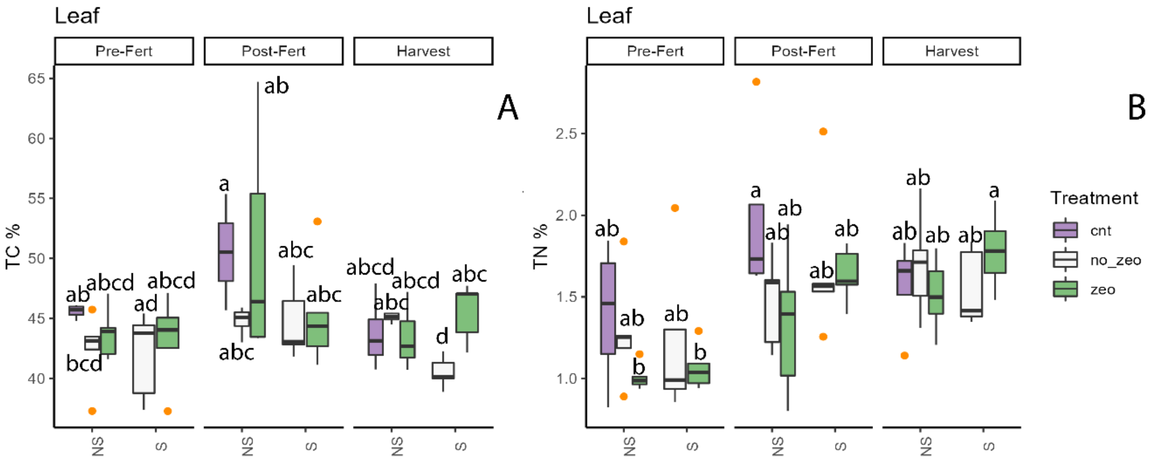

Figure 33. Box-plot of Total Carbon (TC%) (**A**) and Total Nitrogen (TN%) (**B**) in leaves. NS is No-Stress conditions; S means Water Stress. "Pre-Fert" is the first sampling at Pre-Fertilization time; "Post-Fert" represents the middle sampling after Fertilization and "Harvest" is the last sampling at Harvest. Different letters represent significant differences ($p<0.05$) as a result of ANOVA and Tukey (HSD) tests.

Figure 34 did not highlight any difference among treatments in δ^{15}N for both soils and leaves. The ^{15}N natural abundance did not show any particular difference after the fertilization because of the digestate origin. The provenance of digestate from bovine slurry may have affected the plant signature because the digestate applied in the experiment did not have an isotopic signature (8-10‰) as high as digestate from other animal provenance, such as pig slurry (10-20‰ and sometimes even more than >20‰) (Choi et al., 2003, 2006, 2002; Hogberg, 1997; Szpak, 2014). Another consequence could be a too-low N supply in that kind of soil (in terms of the amount of N-input) that, combined with the digestate type, was not enough for the discrimination of different treatments in leaves through the digestate input ($\delta^{15}N$ fingerprint). The $\delta^{15}N$ trend is strongly linked to the Discrimination Factor $\Delta^{13}C$, which represents the normalization of $\delta^{13}C$ for changes in atmospheric CO_2 concentration, by changing photosynthetic rates. No differences were highlighted due to the zeolitite application for both well-watered and stressed plants in $\delta^{15}N$ and $\Delta^{13}C$ values.

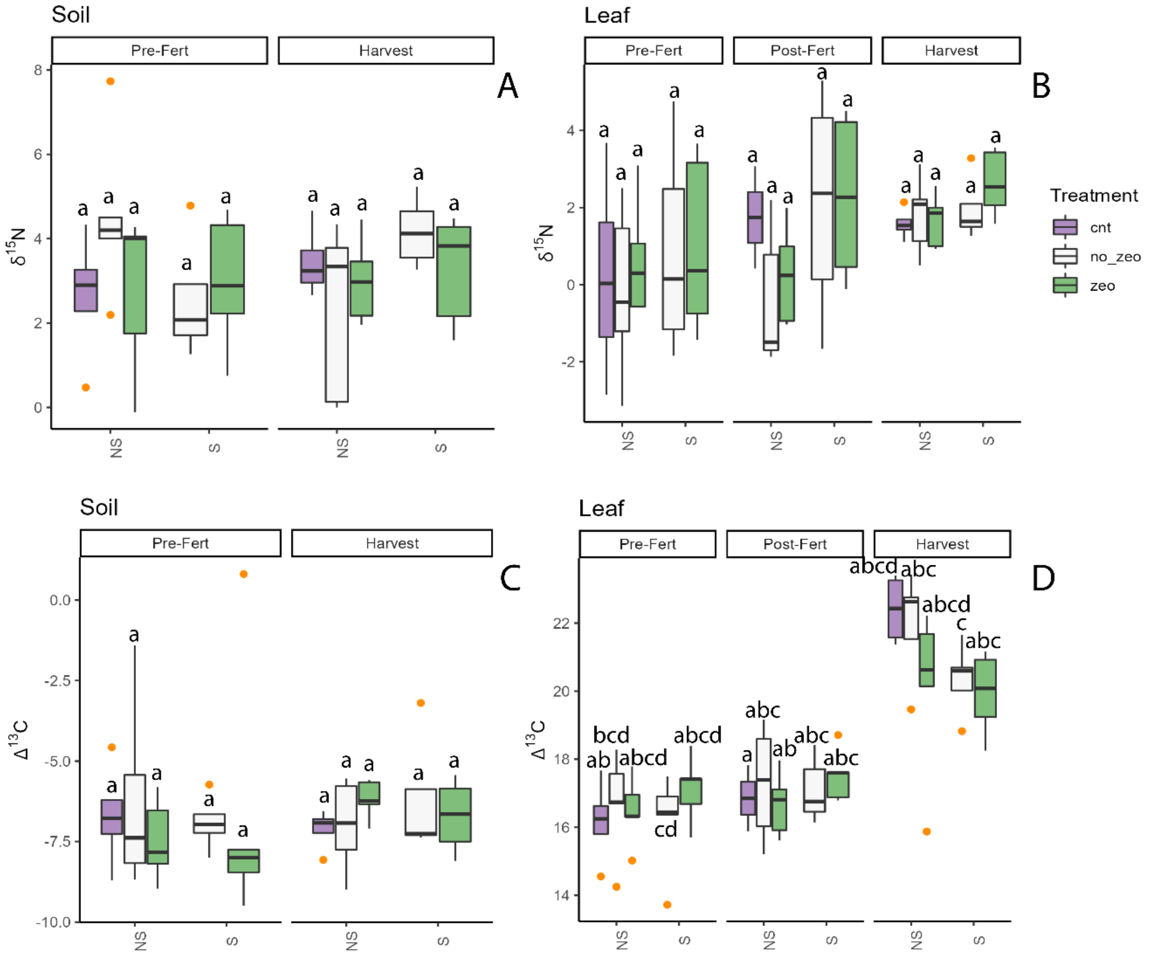

Figure 34. Box-plot of $\delta^{15}N$ in soils (**A**) and leaves (**B**); box-plot of $\Delta^{13}C$ in soils (**C**) and leaves (**D**). NS is No-Stress conditions; S means Water Stress. "Pre-Fert" is the first sampling at Pre-Fertilization time; "Post-Fert" represents the middle sampling after Fertilization and "Harvest" is the last sampling at Harvest. Different letters represent significant differences ($p<0.05$) as a result of ANOVA and Tukey (HSD) tests.

Figure 34D of leaf samples showed a considerable increase in $\Delta^{13}C$ in the last sampling. During the autumn months, the changes in light intensity and rainfall could affect the photosynthesis and consequently, the $\Delta^{13}C$ values of leaves; the exposition of the pot and the random sampling of leaves also could influence this parameter without any discrimination in terms of treatment (as a sort of sampling "error"). However, there are no studies about the responses of the plant to seasonal changes and this makes it difficult to give a complete explanation of what happened. The $\Delta^{13}C$ of leaf samples (Figure 34D) is in the range of 15-25‰ accordingly to the data of pea and barley for a low-medium

δ^{15}N (0-4%) (Maxwell et al., 2014). As reported by Maxwell et al. (2014), Δ^{13}C variations could be explained by a combination of irrigation and fertilization effects. Riehl et al. (2014) for example used a 1‰ Δ^{13}C to distinguish stressed from well-watered plants, without considering the soil fertility effects. In the last sampling, plants subjected to water stress conditions showed a tendency to a lower Δ^{13}C compared to non-stressed plants, accordingly to the data from the literature, but no differences were detected owing to the use of zeolitite. Indeed, the Δ^{13}C is strongly dependent on water stress conditions because a simultaneous decrease in photosynthesis, transpiration and leaf conductance is linked to a shift in the carbon isotope composition (associated with changes in the photosynthetic metabolism of the plant) (Busch et al., 2020; Farquhar et al., 1989; Kumar and Singh, 2009). Figure 35 confirmed these decreasing trends in photosynthesis, evapotranspiration and leaf conductance in water stress conditions.

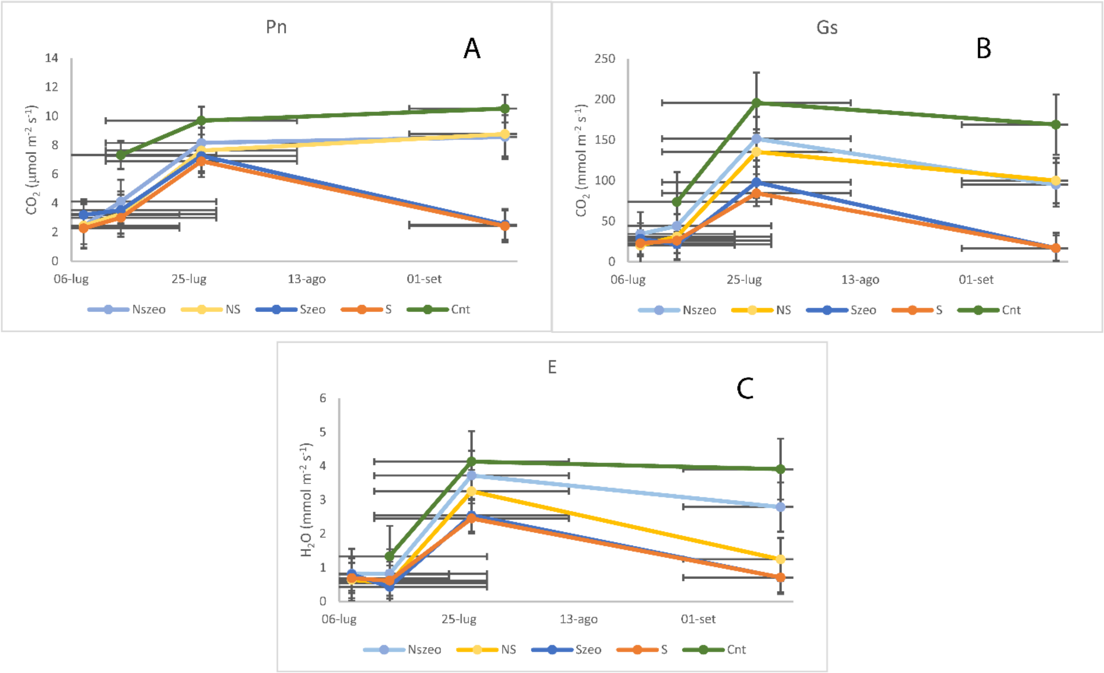

Figure 35. Mean values for (**A**) Net Photoshyntesis (Pn, CO_2 µmol m^{-2} s^{-1}), (**B**) Stomata Conductance (Gs, CO_2 mmol m^{-2} s^{-1}) and (**C**) Transpiration (E, H_2O mmol m^{-2} s^{-1}). Samplings were made on the 14th, 16th, 27th of July and 14th of September. NS is referred to no-stress water conditions, NSzeo is a no-stress water condition and zeolitite, S means water stress, Szeo is water stress and natural zeolitite and Cnt means control.

On the whole, photosynthetic activities showed significantly lower values than those usually measured in grapes (Kizildeniz et al., 2021; Ma et al., 2020). Moreover, the only meaningful difference was found in September when well-irrigated plants (NS and NSzeo) showed better photosynthetic activity than S and Szeo plants (Figure 35A). Gs and E (Figures 35B and C) are linked to the CO_2 fluxes and follow the Pn trend (Figure 35A), but only E highlighted a slight difference in no-stress conditions between grapes with zeolitite (Szeo) and without (S).

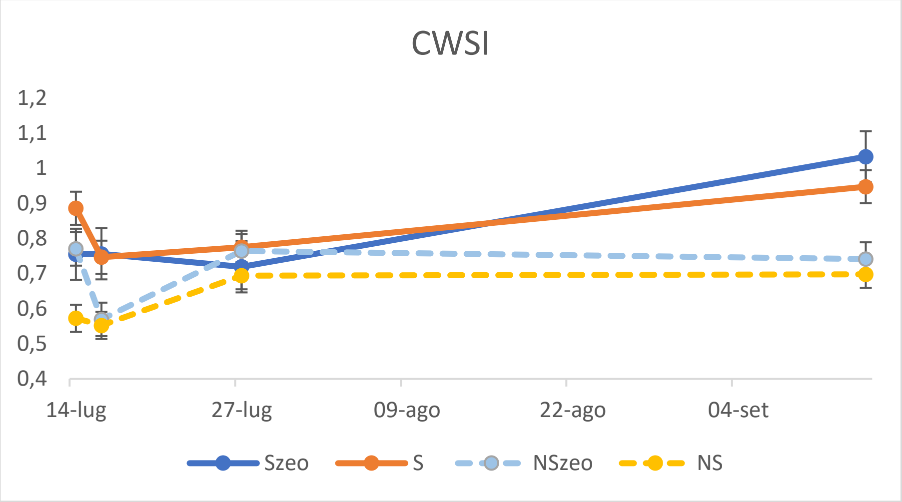

Figure 36. Crop Water Stress Index (CWSI) and std deviation of soil samples. Samplings were made in July (14th, 16th and 27th) and on 14th of September. NS is referred to no-stress water conditions, NSzeo is a no-stress water condition and zeolitite, S means water stress and Szeo is water stress and natural zeolitite.

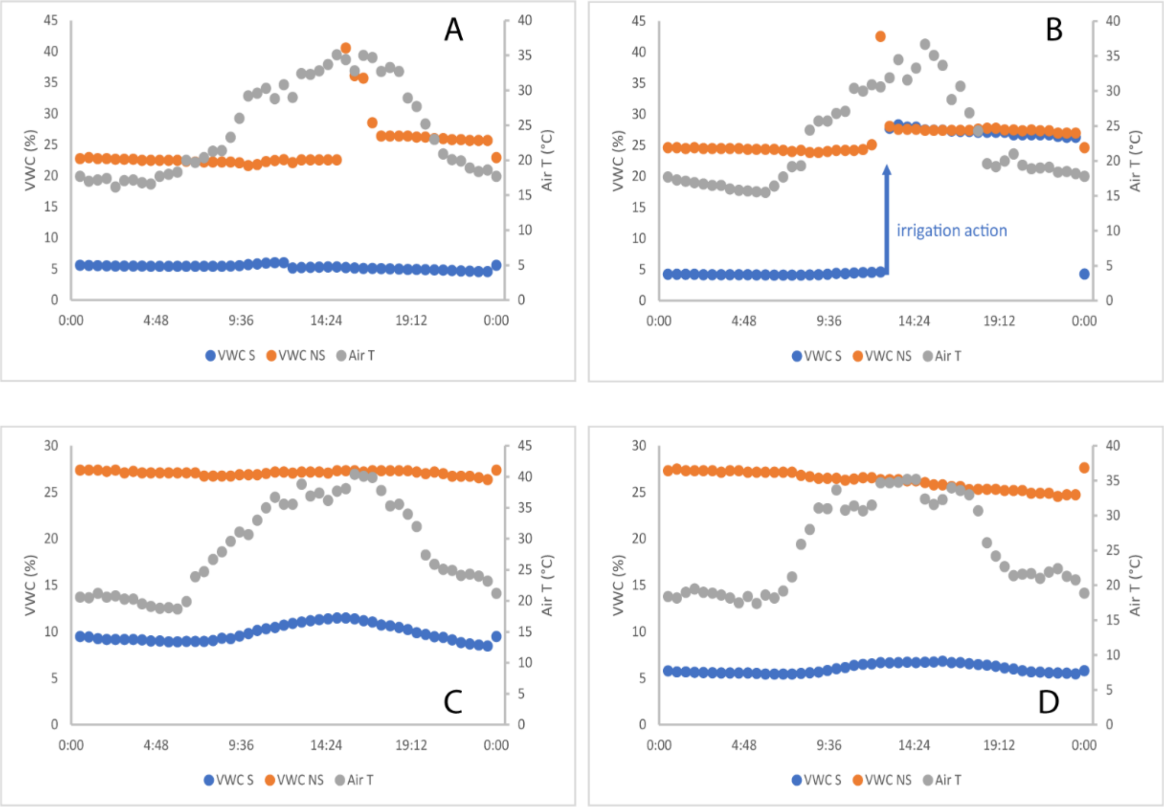

Figure 37. Hourly average values for Volumetric Water Content (VWC%) and air temperature (°C). Samplings were made on 14th July (**A**), 16th July (**B**), 27th July (**C**) and 14th September (**D**). VWC NS is referred to no stress water conditions (in orange), while VWC S means water stress (in blue). Grey points are for air temperature.

The only meaningful differences in CWSI between water stress (S and Szeo) and normal water conditions (NS and NSzeo) appeared in September (Figure 36), while the use of zeolitite did not discriminate different treatments on grapes. However, soil samples with zeolitite showed higher CWSI values than the equivalent sample without zeolitite, although not statistically significant. VWC in Figure 37 well separated the stressed plants against no-stressed plants, except on 16th July when an irrigation action was needed to restore an adequate water content to continue the vegetative activity. This action resulted in similar values between S and NS soil humidity (Figure 37B) after the irrigation. The mean Air Temperatures (Figure 37) reached values above 35°C in the hottest hours, which probably led to a reduced photosynthetic activity. Although CWSI and VWC are usually correlated in soil, from our data no correlation was found, probably due to the i) low Soil Organic Matter, ii) limited time for vegetative recovery and equilibration after the grape potting and iii) no-optimal vegetative status of plants at the beginning of the experiment.

Overall, grapevines were resistant to react to the inputs (such as zeolitite), fertilization and irrigation. In the first months, no meaningful activities were recorded, but some differences were highlighted in September. The early stasis should be due to the non-immediate adjustment time of plants that were transplanted into the new "environment". The no-optimal vegetative status of plants at the beginning could also affect the success of the experiment. Further measurements during the years maybe would have yielded more complete results.

Soil samples (Table 11) highlighted variable chemical composition, with the highest values recorded by Rb and Sr (200-50 ppm), followed by Pr, Nb, Ce, Sm and Zr (50-10 ppm). All other elements showed concentrations below 10 ppm. Rb, Zr, Nb, Pr and Th elements showed significant differences based on the water content, with higher values recorded in normal watering (including CNT) than in water stress conditions. Concerning the presence of zeolite as soil amendment, some elements (Sr, Zr, Nb, La, Ce, Nd, Sm, Eu, Gd, Hf, Th and U) were more concentrated in NSzeo and Szeo with respect to the other treatments without zeolite. In some cases, such as Sr, the values recorded in zeolite treatments were twice that in treatments without zeolite. The application of natural zeolite in soil gave a stronger contribution to the geochemical composition in terms of trace elements and increasing their concentration compared to the treatments without zeolite.

Table 11. Trace elements composition (ppm) of soil samples (mean ± standard deviation). The values are the result of four replicates per treatment. CNT is control, NS means no water stress, NSzeo is no water stress and zeolite in soil, S indicates water stress and Szeo is water stress and zeolite in soil. Different uppercase letters indicate significant differences due to the water stress conditions while lowercase letters indicate significant differences between the treatments with zeolite and without zeolite ($p<0.05$) as a result of ANOVA and Tukey's HSD test.

	CNT		**NS**		**NSzeo**		**S**		**Szeo**	
Rb	61.65A	±14.37	66.22A	±8.65	88.84A	±9.80	50.48B	±10.51	61.08B	±9.06

Sr	93.18b	±11.17	100.55b	±11.83	211.15a	±11.51	70.63b	±9.66	130.71a	±34.15
Zr	9.16Aab	±1.68	10.43Ab	±1.43	16.99Aa	±4.90	6.52Bb	±1.67	10.00Ba	±3.04
Nb	23.94Aab	±4.31	27.92Ab	±3.16	48.75Aa	±11.50	16.81Bb	±4.38	20.87Ba	±3.45
La	6.52ab	±0.75	6.53b	±1.21	8.05a	±0.89	5.85b	±0.45	6.67a	±0.99
Ce	16.94 ab	±2.26	16.40b	±3.10	23.12a	±2.83	16.21b	±2.03	18.27a	±3.32
Pr	36.90A	±6.47	38.36A	±4.76	48.28A	±6.91	24.92B	±3.69	29.50B	±5.80
Nd	3.96 ab	±0.63	3.79b	±0.83	5.14a	±0.69	3.82b	±0.57	4.47a	±0.73
Sm	15.10ab	±2.48	15.67b	±1.99	19.14a	±2.61	14.43b	±1.93	16.89a	±2.68
Eu	2.85ab	±0.50	2.77b	±0.65	3.47a	±0.53	2.70b	±0.29	3.13a	±0.47
Gd	0.57ab	±0.08	0.56b	±0.11	0.71a	±0.08	0.53b	±0.05	0.59a	±0.08
Tb	2.30	±0.45	2.28	±0.56	2.70	±0.45	2.16	±0.21	2.52	±0.34
Dy	0.38	±0.07	0.38	±0.10	0.44	±0.08	0.35	±0.03	0.41	±0.05
Y	1.82	±0.40	1.82	±0.47	2.08	±0.39	1.65	±0.16	1.97	±0.25
Ho	0.36	±0.08	0.37	±0.09	0.42	±0.08	0.32	±0.03	0.38	±0.05
Er	0.96	±0.22	0.96	±0.25	1.09	±0.22	0.86	±0.09	1.03	±0.13
Tm	0.15	±0.03	0.15	±0.04	0.17	±0.04	0.13	±0.01	0.16	±0.02
Yb	0.87	±0.17	0.89	±0.22	1.01	±0.17	0.76	±0.11	0.86	±0.10
Lu	0.13	±0.03	0.13	±0.04	0.15	±0.04	0.12	±0.01	0.14	±0.02
Hf	0.60ab	±0.15	0.60b	±0.21	0.97a	±0.30	0.47b	±0.11	0.67a	±0.18
Ta	0.25	±0.04	0.24	±0.05	0.28	±0.04	0.26	±0.03	0.30	±0.04
Th	6.06Aab	±1.23	6.16Ab	±1.52	10.01Aa	±1.73	4.30Bb	±1.01	5.68Ba	±1.40
U	2.06b	±0.02	2.03b	±0.14	2.82a	±0.11	2.13b	±0.30	2.35a	±0.33

All elements of leaf samples (Table 12) showed significant differences due to the water stress, with the highest values recorded for no water stress conditions. Concerning the presence of zeolite, only Rb showed a meaningful higher value due to zeolite treatments. The more concentrated treatments (NSzeo and Szeo) increased the Rb values by a factor of 5 and 3 than treatments without zeolite (NS and S) in normal water and stress water conditions, respectively.

Table 12. Trace elements composition (ppm) of soil samples (mean ± standard deviation). Values with "*" are expressed in ppb. The values are the result of four replicates per treatment. CNT is control, NS means no water stress, NSzeo is no water stress and zeolite in soil, S indicates water stress and Szeo is water stress and zeolite in soil. Different uppercase letters indicate significant differences due to the water stress conditions while lowercase letters indicate significant differences between the treatments with zeolite and without zeolite ($p<0.05$) as a result of ANOVA and Tukey's HSD test.

	CNT		**NS**		**NSzeo**		**S**		**Szeo**	
Rb	3.892b	±0.937	2.559b	±0.887	13.246a	±3.765	2.991b	±0.898	10.579a	±2.036
Sr	30.721	±11.713	40.407	±11.805	54.158	±10.071	40.762	±14.502	25.278	±9.482
Zr	0.041A	±0.015	0.063A	±0.029	0.096A	±0.078	0.029B	±0.018	0.034B	±0.010
Nb	0.046A	±0.022	0.042A	±0.020	0.079A	±0.041	0.021B	±0.016	0.038B	±0.015
La	0.011A	±0.002	0.020A	±0.018	0.042A	±0.029	0.005B	±0.003	0.010B	±0.007
Ce	0.058A	±0.022	0.086A	±0.036	0.093A	±0.043	0.029B	±0.017	0.043B	±0.013

Pr	0.098A	±0.038	0.145A	±0.064	0.166A	±0.072	0.049B	±0.027	0.077B	±0.026
Nd	0.013A	±0.005	0.019A	±0.009	0.022A	±0.009	0.006B	±0.004	0.010B	±0.004
Sm	0.051A	±0.021	0.077A	±0.035	0.087A	±0.036	0.026B	±0.014	0.041B	±0.014
Eu	0.010A	±0.004	0.015A	±0.007	0.017A	±0.007	0.007B	±0.005	0.008B	±0.003
Gd	0.003A	±0.001	0.004A	±0.002	0.004A	±0.001	0.002B	±0.001	0.002B	±0.001
Tb	0.009A	±0.004	0.013A	±0.007	0.014A	±0.005	0.005B	±0.003	0.007B	±0.002
Dy	0.001A	±0.001	0.002A	±0.001	0.002A	±0.001	*0.70B	±0.000	0.001B	±0.000
Y	0.006A	±0.003	0.010A	±0.005	0.010A	±0.004	0.005B	±0.003	0.005B	±0.001
Ho	0.001A	±0.000	0.002A	±0.001	0.002A	±0.001	*0.60B	±0.000	0.001B	±0.000
Er	0.003A	±0.001	0.005A	±0.002	0.005A	±0.002	0.002B	±0.001	0.003B	±0.001
Tm	*0.50A	±0.000	0.001A	±0.000	0.001A	±0.000	*0.30B	±0.000	*0.30B	±0.000
Yb	0.003A	±0.001	0.004A	±0.002	0.004A	±0.002	0.002B	±0.001	0.002B	±0.001
Lu	*0.40A	±0.000	0.001A	±0.000	0.001A	±0.000	*0.20B	±0.000	*0.30B	±0.000
Hf	0.002A	±0.000	0.002A	±0.001	0.003A	±0.001	0.001B	±0.001	0.001B	±0.001
Ta	0.006Aa	±0.002	0.003Ab	±0.001	0.003Ab	±0.001	0.001Bb	±0.000	0.002Bb	±0.001
Th	0.024A	±0.008	0.035A	±0.013	0.028A	±0.014	0.010B	±0.004	0.018B	±0.007
U	0.005A	±0.002	0.007A	±0.002	0.011A	±0.008	0.003B	±0.002	0.004B	±0.002

PCA of soils (Figure 38A) indicated 89.29% of the total variance, divided in 77.44% of PC1 and 11.85% of PC2. NSzeo treatments was more separated from the other treatments, also according to the PLS-DA analysis showed in Figure 38C. Szeo was well separated from CNT, S and NS treatments (Figure 38C), which means that the use of zeolite as soil amendment was a discriminator variable when studying trace elements. Lower differences were found in the PLS-component t2 for the water stress conditions. VIP analysis (Figure 38E) highlighted Sr, Ce, Pr, Ho, Er, Yb, Ta and U as the discriminator for soil samples.

PCA of leaves indicated 82.36% of PC1 and 6.45% of PC2, with 88.81% of the total variance (Figure 38B). The PLS-DA (Figure 38D) highlighted a meaningful separation between treatments with zeolite and other treatments without (PLS-component t1 in Figure 38D), while a lower separation in the secondary component (PLS-component t2) allows to distinguish leaf samples due to the water stress. VIP analysis (Figure 38F) pointed out Rb as the best discriminant element, followed by Sr and Ta.

Overall, both soils and leaves showed a significant separation between the treatments with zeolitite (NSzeo and Szeo) and the unamended treatments (CNT, NS and S), probably thanks to the impact of zeolite minerals on the soil geochemistry. The contribution of zeolites in terms of trace elements also occurred in leaf samples, probably due to the plant uptake from soils. The most discriminating elements from the comparison of soil and leaf in VIP analysis were Rb, Sr and Ta (Figures 38E and F). In order to verify the capability of plants to uptake chemical elements from soil, the index of Bioaccumulation (BA) was calculated following Equation 2.

BA coefficient confirmed the results of PCA and PLS-DA in terms of water stress conditions, showing that plant uptake was more efficient in normal water than in stressed water conditions (Table 12). Concerning the use of zeolitite, Rb was the only element showing a meaningful difference in the plant uptake thanks to the zeolitite, which probably means that zeolites minerals help soil to retain and slowly release water available to the plant. Nevertheless, in water stress conditions almost all elements highlighted a higher trend for Szeo than S, although not significant. In normal water conditions only Rb, Nb, La, Ce, Pr, Gd, Y, Tm and Hf showed the same trend, while for all other elements the NS had a higher BA coefficient than NSzeo.

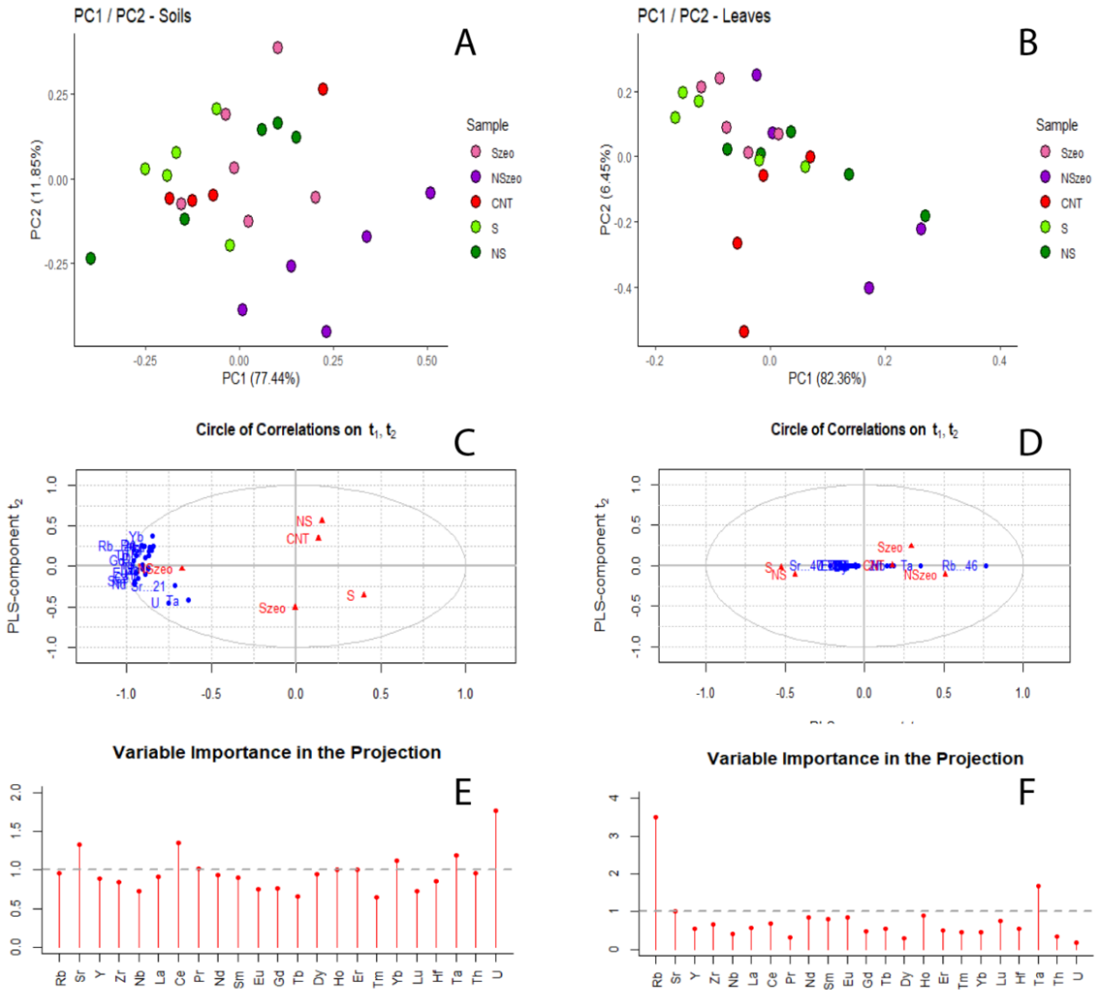

Figure 38. Soil and leaf samples: (**A**) and (**B**) PCA score plot, (**C**) and (**D**) PLS-DA score plot and (**E**) and (**F**) Variable Importance in the Projection (VIP). CNT is the control treatment, NS is referred to no stress water conditions, NSzeo is no-stress water conditions and zeolitite, S means water stress and Szeo is water stress and natural zeolitite. Observation axes PC1 and PC2: 89.29% for soils and 88.81% for leaves.

Table 12. Bioaccumulation Coefficient (BA). CNT is the control treatment, NS is referred to no-stress water conditions, NSzeo is no-stress water conditions and zeolitite, S means water stress and Szeo is water stress and natural zeolitite. Different uppercase letters indicate significant differences due to the water stress conditions while lowercase letters indicate significant differences between the treatments with zeolite and without zeolite ($p<0.05$) as a result of ANOVA and Tukey's HSD test.

	CNT		NS		NSzeo		S		Szeo	
Rb	0,029ab	±0,010	0,017b	±0,004	0,058a	±0,026	0,026b	±0,008	0,076a	±0,010
Sr	0,045ab	±0,012	0,057a	±0,019	0,032b	±0,011	0,065a	±0,033	0,030b	±0,010
Zr	0,001	±0,000	0,001	±0,000	0,001	±0,001	0,001	±0,001	0,001	±0,001
Nb	0,002	±0,001	0,002	±0,001	0,004	±0,004	0,003	±0,003	0,002	±0,001
La	0,038A	±0,013	0,045A	±0,026	0,050A	±0,021	0,028B	±0,018	0,027B	±0,006
Ce	0,009A	±0,003	0,010A	±0,001	0,011A	±0,004	0,005B	±0,002	0,007B	±0,002
Pr	0,004	±0,001	0,004	±0,003	0,005	±0,002	0,003	±0,001	0,004	±0,001
Nd	0,023A	±0,006	0,051A	±0,036	0,036A	±0,013	0,015B	±0,008	0,020B	±0,006
Sm	0,005A	±0,002	0,008A	±0,006	0,006A	±0,002	0,002B	±0,001	0,003B	±0,001
Eu	0,018A	±0,006	0,030A	±0,020	0,020A	±0,007	0,013B	±0,005	0,012B	±0,004
Gd	0,063A	±0,023	0,078A	±0,063	0,099A	±0,036	0,043B	±0,023	0,062B	±0,021
Tb	0,017A	±0,008	0,031A	±0,024	0,024A	±0,008	0,009B	±0,005	0,013B	±0,004
Dy	0,054A	±0,021	0,125A	±0,102	0,094A	±0,037	0,038B	±0,018	0,052B	±0,016
Y	0,015A	±0,006	0,018A	±0,011	0,019A	±0,008	0,008B	±0,002	0,011B	±0,003
Ho	0,050A	±0,021	0,110A	±0,088	0,085A	±0,032	0,035B	±0,016	0,049B	±0,018
Er	0,016A	±0,008	0,036A	±0,029	0,027A	±0,011	0,012B	±0,005	0,015B	±0,005
Tm	0,134A	±0,074	0,122A	±0,100	0,162A	±0,070	0,086B	±0,049	0,111B	±0,039
Yb	0,021A	±0,011	0,033A	±0,024	0,022A	±0,009	0,011B	±0,005	0,015B	±0,005
Lu	0,152A	±0,053	0,210A	±0,172	0,150A	±0,066	0,066B	±0,033	0,094B	±0,036
Hf	0,027	±0,006	0,019	±0,012	0,030	±0,015	0,026	±0,015	0,022	±0,010
Ta	1,397Aa	±0,161	1,041Ab	±0,456	0,642Ab	±0,115	0,337Bb	±0,116	0,557Bb	±0,194
Th	0,144	±0,063	0,203	±0,156	0,094	±0,036	0,073	±0,026	0,114	±0,054
U	0,264A	±0,111	0,422A	±0,208	0,328A	±0,186	0,201B	±0,118	0,176B	±0,040

7. Conclusions of grapevine experiment

Thanks to the monitoring of a 2-years grapevine *Cv* Glera, the influence of different agricultural practices was tested in a shade canopy environment.

The water stress conditions mainly affected the plant's physiological status and TC in leaves, given that irrigation influences plant growth and photosynthesis. As a direct consequence of the water condition, also a small variation in $\Delta^{13}C$ allowed to discriminate between well-irrigated and no-irrigated plants.

Regarding the zeolitite effects, the best results were related to the geochemical composition of soils and leaves. Some trace elements were more concentrated in soils amended with zeolitite than no-amended soils, proving that zeolitite provided some elements to the soil. For example, Sr in zeolitite treatments (NSzeo and Szeo) was twice than in NS and S treatments. In leaf samples, only Rb was meaningfully higher in zeolitite treatments than others. Moreover, BA coefficient highlighted that almost all elements, especially Rb, were higher in zeolitite treatment than in no-zeolitite treatment in water stress conditions, while normal water conditions in leaves showed no differences between treatments. Statistical analyses, PCA and PLS-DA, apparently confirmed the importance of zeolitite in the treatment discrimination for both soils and leaves, while water conditions had less separation among all treatments. VIP analysis highlighted Rb, Sr and Ta as the best discriminator elements of the zeolitite use.

Thereby, the use of different agricultural practices, mainly the use of zeolitite as soil amendment and the water condition, led to a meaningful alteration of some plant parameters and to the geochemical signature in soils and leaves. Of course, more studies are required in order to investigate the geochemical and isotopic fingerprint in grapes, which was not possible due to the too-young grapevine of the experiment.

8. General remarks

The PhD research was focused on the investigation of innovative methods and eco-friendly strategies in agricultural practices through the application of zeolitite, that allowed to i) improve the low efficiency of fertilizers and chemical products and therefore ii) decrease their overuse in agriculture. In addition, the geochemical and isotopic compositions of Agri-Food products were investigated to guarantee their traceability according to geographical origin and agricultural practices.

The importance of zeolitite as soil amendment to retain and slowly release nutrients and water, a well-known and applied technique in many crops, was demonstrated in olive growing and grapevine. The use of zeolitite as soil improver in olive growing allowed to reduce 50% of N-input from fertilization, reducing the N losses in the environment without effects on plant growth. Zeolitite is likely helping grapevine photosynthesis in water stress conditions, in terms of Total Carbon, and its evapotranspiration in normal water conditions.

In terms of food traceability, the geochemical and isotopic composition in plants and final products coupled with different soil types and climatic features (*Terroir*) allowed to discriminate olives and olive oils according to their geographical origin. Moreover, the foliar treatment fingerprint as a tool for identifying Agri-Food products represented the most innovative aspect of the traceability study. Therefore, the management of different agricultural practices could establish the groundwork to develop a geochemical "branding" in final products.

This PhD research presented interesting points for a discerning and thorough investigation on food traceability and agricultural practices sustainability. However, further studies, such as testing different types of geomaterials or more crops, would help boosting the agricultural sector innovation.

9. Acknowledgements

I would like to thank my supervisor Prof. Massimo Coltorti and the co-supervisor Dr. Giacomo Ferretti for their insightful comments and suggestions during my PhD study. I also am really grateful to my family for the encouragement in the past years and to Luca for the emotional support and his endless patience.

10. Bibliography

Abreu, M.M., Santos, E.S., Magalhães, M.C.F., Fernandes, E., 2012. Trace elements tolerance, accumulation and translocation in Cistus populifolius, Cistus salviifolius and their hybrid growing in polymetallic contaminated mine areas. J. Geochemical Explor. 123, 52–60. https://doi.org/10.1016/J.GEXPLO.2012.05.001

Aceto, M., Calà, E., Musso, D., Regalli, N., Oddone, M., 2019. A preliminary study on the authentication and traceability of extra virgin olive oil made from Taggiasca olives by means of trace and ultra-trace elements distribution. Food Chem. 298. https://doi.org/10.1016/J.FOODCHEM.2019.125047

Ackman, R.G., 2002. The gas chromatograph in practical analyses of common and uncommon fatty acids for the 21st century. Anal. Chim. Acta 465, 175–192. https://doi.org/10.1016/S0003-2670(02)00098-3

Agnan, Y., Séjalon-Delmas, N., Probst, A., 2014. Origin and distribution of rare earth elements in various lichen and moss species over the last century in France. Sci. Total Environ. 487, 1–12. https://doi.org/10.1016/J.SCITOTENV.2014.03.132

Al-Bachir, M., Sahloul, H., 2017. Fatty acid profile of olive oil extracted from irradiated and non-irradiated olive fruits. Int. J. Food Prop. https://doi.org/10.1080/10942912.2016.1243557

Alagić, S., Tošić, S.B., Dimitrijević, M.D., Antonijević, M.M., Nujkić, M.M., 2015. Assessment of the quality of polluted areas based on the content of heavy metals in different organs of the grapevine (Vitis vinifera) cv Tamjanika. Environ. Sci. Pollut. Res. 22, 7155–7175. https://doi.org/10.1007/S11356-014-3933-1/FIGURES/9

Alboukadel Kassambara, 2020. ggpubr: ‘ggplot2’ Based Publication Ready Plots. R package version 0.4.0.

Almeida, C.M.R., Vasconcelos, M.T.S.D., 2003. Multielement composition of wines and their precursors including provenance soil and their potentialities as fingerprints of wine origin. J. Agric. Food Chem. 51, 4788–4798. https://doi.org/10.1021/JF034145B/ASSET/IMAGES/LARGE/JF034145BF00007.JPEG

American Oil Chemists’ Society, AOCS Official Method, 2006a.

American Oil Chemists’ Society, AOCS Official Method, 2006b. Ce 1c-89.

Amorós, J.A., Pérez-de-los Reyes, C., García Navarro, F.J., Bravo, S., Chacón, J.L., Martínez, J., Jiménez Ballesta, R., 2013. Bioaccumulation of mineral elements in grapevine varieties cultivated in ‘La Mancha’. J. Plant Nutr. Soil Sci. 176, 843–850. https://doi.org/10.1002/JPLN.201300015

Amundson, R., Austin, A.T., Schuur, E.A.G., Yoo, K., Matzek, V., Kendall, C., Uebersax, A., Brenner, D., Baisden, W.T., 2003. Global patterns of the isotopic composition of soil and plant nitrogen. Global Biogeochem. Cycles 17, 1031. https://doi.org/10.1029/2002GB001903

Angelova, V.R., Ivanov2, A.S., Braikov2, D.M., 1999. Heavy metals (Pb, Cu, Zn and Cd) in the system soil-grapevine-grape. J. Sci. Food Agric. J Sci Food Agric 79, 713–721. https://doi.org/10.1002/(SICI)1097-0010(199904)79:5

Ayton, J., Mailer, R.J., Haigh, A., Tronson, D., Conlan, D., 2007. QUALITY AND OXIDATIVE STABILITY OF AUSTRALIAN OLIVE OIL ACCORDING TO HARVEST DATE AND IRRIGATION. J. Food Lipids 14, 138–156. https://doi.org/10.1111/J.1745-4522.2007.00076.X

Baccouri, B., Rajhi, I., Mohamed, S.N., Flamini, G., 2022. Monitoring the fatty acid and volatile compositions of Tunisian virgin olive oils using HS-SPME–GC–MS with regard to growing area. Eur. Food Res. Technol. 248, 2877–2885. https://doi.org/10.1007/S00217-022-04097-6/FIGURES/5

Barker, M., Rayens, W., 2003. Partial least squares for discrimination. J Chemom. 17, 166–73. https://doi.org/10.1002/cem.785

Barret Schloerke, Di Cook, Joseph Larmarange, Francois Briatte, M., Marbach, Edwin Thoen, A.E. and J.C., 2021. GGally: Extension to 'ggplot2.

Basso, B., Dumont, B., Cammarano, D., Pezzuolo, A., Marinello, F., Sartori, L., 2016. Environmental and economic benefits of variable rate nitrogen fertilization in a nitrate vulnerable zone. Sci. Total Environ. 545, 227–235. https://doi.org/10.1016/j.scitotenv.2015.12.104

Bateman, A.S., Kelly, S.D., 2007. Fertilizer nitrogen isotope signatures. Isotopes Environ. Health Stud. 43, 237–247. https://doi.org/10.1080/10256010701550732

Baxter, M.J., Crews, H.M., Dennis, M.J., Goodall, I., Anderson, D., 1997. The determination of the authenticity of wine from its trace element composition. Food Chem. 60, 443–450. https://doi.org/10.1016/S0308-8146(96)00365-2

Bell, S.-J., Henschke, P.A., 2005. Implications of nitrogen nutrition for grapes, fermentation and wine. Aust. J. Grape Wine Res. 11, 242–295. https://doi.org/10.1111/j.1755-0238.2005.tb00028.x

Benincasa, C., Lewis, J., Perri, E., Sindona, G., Tagarelli, A., 2007. Determination of trace element in Italian virgin olive oils and their characterization according to geographical origin by statistical analysis. Anal. Chim. Acta 585, 366–370. https://doi.org/10.1016/J.ACA.2006.12.040

Benincasa, C., Lewis, J., Sindona, G., Tagarelli, A., 2008. The use of multi element profiling to differentiate between cowand buffalo milk. Food Chem. 110, 257–262. https://doi.org/10.1016/J.FOODCHEM.2008.01.049

Bensmira, M., Jiang, B., Nsabimana, C., Jian, T., 2007. Effect of Lavender and Thyme incorporation in sunflower seed oil on its resistance to frying temperatures. undefined 40, 341–346. https://doi.org/10.1016/J.FOODRES.2006.10.004

Boggia, R., Zunin, P., Lanteri, S., Rossi, N., Evangelisti, F., 2002. Classification and class-modeling of 'Riviera Ligure' extra-virgin olive oil using chemical-physical parameters. J. Agric. Food Chem. 50, 2444–2449. https://doi.org/10.1021/JF011289M/ASSET/IMAGES/LARGE/JF011289MF00004.JPEG

Bontempo, L., Camin, F., Larcher, R., Nicolini, G., Perini, M., Rossmann, A., 2009. Coast and year effect on H, O and C stable isotope ratios of Tyrrhenian and Adriatic italian olive oils. Rapid Commun. Mass Spectrom. 23, 1043–1048. https://doi.org/10.1002/rcm.3968

Borges, T.H., Pereira, J.A., Cabrera-Vique, C., Lara, L., Oliveira, A.F., Seiquer, I., 2017. Characterization of Arbequina virgin olive oils produced in different regions of Brazil and

Spain: Physicochemical properties, oxidative stability and fatty acid profile. Food Chem. 215, 454–462. https://doi.org/10.1016/J.FOODCHEM.2016.07.162

Bravo, S., Amorós, J.A., Pérez-de-los-Reyes, C., García, F.J., Moreno, M.M., Sánchez-Ormeño, M., Higueras, P., 2017. Influence of the soil pH in the uptake and bioaccumulation of heavy metals (Fe, Zn, Cu, Pb and Mn) and other elements (Ca, K, Al, Sr and Ba) in vine leaves, Castilla-La Mancha (Spain). J. Geochemical Explor. 174, 79–83. https://doi.org/10.1016/J.GEXPLO.2015.12.012

Bremmer, J., Gonzalez-Martinez, A., Jongeneel, R., Huiting, H., Stokkers, R., Ruijs, M., 2021. Impact assessment of EC 2030 Green Deal Targets for sustainable crop production. Wageningen, The Netherlands. https://doi.org/10.18174/558517

Brescia, M.A., Monfreda, M., Buccolieri, A., Carrino, C., 2005. Characterisation of the geographical origin of buffalo milk and mozzarella cheese by means of analytical and spectroscopic determinations. Food Chem. 89, 139–147. https://doi.org/10.1016/J.FOODCHEM.2004.02.016

Brioschi, L., Steinmann, M., Lucot, E., Pierret, M.C., Stille, P., Prunier, J., Badot, P.M., 2013. Transfer of rare earth elements (REE) from natural soil to plant systems: Implications for the environmental availability of anthropogenic REE. Plant Soil 366, 143–163. https://doi.org/10.1007/S11104-012-1407-0/FIGURES/9

Brondz, I., 2002. Development of fatty acid analysis by high-performance liquid chromatography, gas chromatography, and related techniques. Anal. Chim. Acta 465, 1–37. https://doi.org/10.1016/S0003-2670(01)01467-2

Buscaroli, A., 2017. An overview of indexes to evaluate terrestrial plants for phytoremediation purposes (Review). Ecol. Indic. 82, 367–380. https://doi.org/10.1016/J.ECOLIND.2017.07.003

Busch, F.A., Holloway-Phillips, M., Stuart-Williams, H., Farquhar, G.D., 2020. Revisiting carbon isotope discrimination in C3 plants shows respiration rules when photosynthesis is low. Nat. Plants 2020 63 6, 245–258. https://doi.org/10.1038/s41477-020-0606-6

Campisi, T., Abbondanzi, F., Faccini, B., Di Giuseppe, D., Malferrari, D., Coltorti, M., Laurora, A., Passaglia, E., 2016. Ammonium-charged zeolitite effects on crop growth and nutrient leaching: Greenhouse experiments on maize (Zea mays). Catena 140, 66–76. https://doi.org/10.1016/j.catena.2016.01.019

Cantrell, K.J., Byrne, R.H., 1987. Rare earth element complexation by carbonate and oxalate ions. Geochim. Cosmochim. Acta 51, 597–605. https://doi.org/10.1016/0016-7037(87)90072-X

Capo, R.C., Stewart, B.W., Chadwick, O.A., 1998. Strontium isotopes as tracers of ecosystem processes: theory and methods. Geoderma 82, 197–225. https://doi.org/10.1016/S0016-7061(97)00102-X

Cataldo, E., Salvi, L., Paoli, F., Fucile, M., Masciandaro, G., Manzi, D., Masini, C.M., Mattii, G.B., 2021. Application of Zeolites in Agriculture and Other Potential Uses: A Review. Agron. 2021, Vol. 11, Page 1547 11, 1547. https://doi.org/10.3390/AGRONOMY11081547

Censi, P., Saiano, F., Pisciotta, A., Tuzzolino, N., 2014. Geochemical behaviour of rare earths in Vitis vinifera grafted onto different rootstocks and growing on several soils. Sci. Total Environ. 473–474, 597–608. https://doi.org/10.1016/J.SCITOTENV.2013.12.073

Cheary, R.W., Coelho, A.A., Cline, J.P., 2004. Fundamental Parameters Line Profile Fitting in Laboratory Diffractometers. J. Res. Natl. Inst. Stand. Technol. 109, 1. https://doi.org/10.6028/JRES.109.002

Chien, S.H., Teixeira, L.A., Cantarella, H., Rehm, G.W., Grant, C.A., Gearhart, M.M., 2016. Agronomic Effectiveness of Granular Nitrogen/Phosphorus Fertilizers Containing Elemental Sulfur with and without Ammonium Sulfate: A Review. Agron. J. 108, 1203–1213.

https://doi.org/10.2134/AGRONJ2015.0276

Choi, W.-J., Ro, H.-M., Hobbie, E.A., 2003. Patterns of natural 15N in soils and plants from chemically and organically fertilized uplands. Soil Biol. Biochem. 35, 1493–1500. https://doi.org/10.1016/S0038-0717(03)00246-3

Choi, W.J., Arshad, M.A., Chang, S.X., Kim, T.H., 2006. Grain 15N of crops applied with organic and chemical fertilizers in a four-year rotation. Plant Soil 284, 165–174. https://doi.org/10.1007/s11104-006-0038-8

Choi, W.J., Lee, S.M., Ro, H.M., Kim, K.C., Yoo, S.H., 2002. Natural 15N abundances of maize and soil amended with urea and composted pig manure. Plant Soil 245, 223–232. https://doi.org/10.1023/A:1020475017254

Choo, L.N.L.K., Ahmed, O.H., Talib, S.A.A., Ghani, M.Z.A., Sekot, S., 2020. Clinoptilolite zeolite on tropical peat soils nutrient, growth, fruit quality, and yield of carica papaya L. CV. sekaki. Agronomy 10. https://doi.org/10.3390/AGRONOMY10091320

Christopher H. Evans, 1990. Biochemistry of the Lanthanides.

Christy, A.A., Egeberg, P.K., 2006. Quantitative determination of saturated and unsaturated fatty acids in edible oils by infrared spectroscopy and chemometrics. Chemom. Intell. Lab. Syst. 82, 130–136. https://doi.org/10.1016/J.CHEMOLAB.2005.06.019

Chua, H., 1998. Bio-accumulation of environmental residues of rare earth elements in aquatic flora Eichhornia crassipes (Mart.) Solms in Guangdong Province of China. Sci. Total Environ. 214, 79–85. https://doi.org/10.1016/S0048-9697(98)00055-2

Colombani, N., Di Giuseppe, D., Faccini, B., Ferretti, G., Mastrocicco, M., Coltorti, M., 2016. Estimated Water Savings in an Agricultural Field Amended With Natural Zeolites. Environ. Process. 3, 617–628. https://doi.org/10.1007/s40710-016-0151-5

Colombani, N., Mastrocicco, M., Di Giuseppe, D., Faccini, B., Coltorti, M., 2015. Batch and column experiments on nutrient leaching in soils amended with Italian natural zeolitites. Catena 127, 64–71. https://doi.org/10.1016/j.catena.2014.12.022

Coltorti, M., Di Giuseppe, D., Faccini, B., Passaglia, E., Malferrari, D., Mastrocicco, M., Colombani, N., 2012. ZeoLIFE, a project for water pollution reduction and water saving using a natural zeolitite cycle. Rend. Online Soc. Geol. Ital. 21, 853.

Commission on Natural Zeolites, n.d. www.iza-online.org/natural/Datasheets [WWW Document].

Concas, S., Lattanzi, P., Bacchetta, G., Barbafieri, M., Vacca, A., 2015. Zn, Pb and Hg contents of Pistacia lentiscus L. grown on heavy metal-rich soils: Implications for phytostabilization. Water. Air. Soil Pollut. 226, 1–15. https://doi.org/10.1007/S11270-015-2609-X/FIGURES/5

Coombs, D.S., 2001. Recommended nomenclature for zeolite minerals: Report of the subcommittee on zeolites of the international mineralogical association, commission on new minerals and mineral names. Bull. Mineral. Petrol. Geochemistry 20, 149–155.

Cugnetto, A., Santagostini, L., Rolle, L., Guidoni, S., Gerbi, V., Novello, V., 2014. Tracing the “terroirs” via the elemental composition of leaves, grapes and derived wines in cv Nebbiolo (Vitis vinifera L.). Sci. Hortic. (Amsterdam). 172, 101–108. https://doi.org/10.1016/J.SCIENTA.2014.03.055

D’Imperio, M., Dugo, G., Alfa, M., Mannina, L., Segre, A.L., 2007a. Statistical analysis on Sicilian olive oils. Food Chem. 102, 956–965. https://doi.org/10.1016/J.FOODCHEM.2006.03.003

D’Imperio, M., Mannina, L., Capitani, D., Bidet, O., Rossi, E., Bucarelli, F.M., Quaglia, G.B., Segre, A., 2007b. NMR and statistical study of olive oils from Lazio: A geographical, ecological and agronomic characterization. Food Chem. 105, 1256–1267. https://doi.org/10.1016/J.FOODCHEM.2007.02.045

Dawson, J.C., Huggins, D.R., Jones, S.S., 2008. Characterizing nitrogen use efficiency in natural and agricultural ecosystems to improve the performance of cereal crops in low-input and organic agricultural systems. F. Crop. Res. 107, 89–101. https://doi.org/10.1016/j.fcr.2008.01.001

De França, E.J., De Nadai Fernandes, E.A., Turra, C., Bacchi, M.A., Elias, C., Tagliaferro, F.S., Moreira, C.F., Ribeiro, A.P., Figueira, R.C.L., 2011. Survey of lanthanoids in plants from a tropical region. Int. J. Environ. Heal. 5, 32–48. https://doi.org/10.1504/IJENVH.2011.039854

De la Guardia, M., Garrigues, S., 2015. Handbook of Mineral Elements in Food. 803.

De Mendiburu, F., 2014. Agricolae: Statistical procedures for agricultural research. R package version 1.2-0. http://CRAN.R-project.org/package=agricolae.

De Smedt, C., Someus, E., Spanoghe, P., 2015. Potential and actual uses of zeolites in crop protection. Pest Manag. Sci. https://doi.org/10.1002/ps.3999

De Smedt, C., Steppe, K., Spanoghe, P., 2017. Beneficial effects of zeolites on plant photosynthesis. Adv. Mater. Sci. 2, 1–11. https://doi.org/10.15761/ams.1000115

de Wit, M., Hugo, A., Shongwe, N., van der Merwe, R., 2016. Effect of cultivar, season and locality on lipid content and fatty acid composition of cactus pear seed oil. South African J. Plant Soil 33, 279–288. https://doi.org/10.1080/02571862.2016.1141335

Delkash, M., Ebrazi Bakhshayesh, B., Kazemian, H., 2015. Using zeolitic adsorbents to cleanup special wastewater streams: A review. Microporous Mesoporous Mater. 214, 224–241. https://doi.org/10.1016/j.micromeso.2015.04.039

Di Giuseppe, D., Ferretti, G., Faccini, B., Blasi, E., Passeri, N., Bianchini, G., Coltorti, M., 2016. Is it possible to cultivate corn in a sustainable way using a quarry waste? Period. di Mineral. 85, 179–183. https://doi.org/10.2451/2016PM633

Di Vaio, C., Graziani, G., Gaspari, A., De Luca, L., Aiello, A., Cirillo, A., Bruno, A., Romano, R., Ritieni, A., 2021. Drupe Characters, Fatty Acids, Polyphenolic and Aromatic Profile of Olive Oil Obtained from “Oliva Bianca”, Minor Autochthonous Cultivar of Campania. Plants 2021, Vol. 10, Page 1119 10, 1119. https://doi.org/10.3390/PLANTS10061119

Díaz, L., Borges, M.E., 2012. Low-quality vegetable oils as feedstock for biodiesel production using k-pumice as solid catalyst. Tolerance of water and free fatty acids contents. J. Agric. Food Chem. 60, 7928–7933. https://doi.org/10.1021/JF301886D/ASSET/IMAGES/LARGE/JF-2012-01886D_0003.JPEG

Dinov, I.D., 2018. Data Science and Predictive Analytics, Data Science and Predictive Analytics. https://doi.org/10.1007/978-3-319-72347-1

Doebelin, N., Kleeberg, R., 2015. Profex: a graphical user interface for the Rietveld refinement program BGMN. urn:issn:1600-5767 48, 1573–1580. https://doi.org/10.1107/S1600576715014685

Drechsel, P., Heffer, P., Magen, H., Mikkelsen, R., Wichelns, D., 2015. Managing Water and Fertilizer for Sustainable Agricultural Intensification, International Fertilizer Industry Association. IFA, IWMI, IPNI and IPI, Paris.

Drivelos, S.A., Georgiou, C.A., 2012. Multi-element and multi-isotope-ratio analysis to determine the geographical origin of foods in the European Union. TrAC - Trends Anal. Chem. 40, 38–51. https://doi.org/10.1016/J.TRAC.2012.08.003

Dukov, I.L., 2007. Nomenclature of inorganic chemistry - IUPAC recommendations 2005. Chemistry (Easton). 16, 561–568. https://doi.org/10.1515/CI.2005.27.6.25/PDF

Echtebarne, F.; Ojeda, H.; H.J.J., 2010. Leaf: fruit ratio and vine water status effects on grenache noir (Vitis vinifera L.) berry composition: water, sugar, organic acids and cations. S. Afr. J. Enol. Vitic. 33, 106–115.

El-Sherpiny, M., Baddour, A.G., El-Kafrawy, M.M., 2020. Effect of Zeolite Soil addition under Different Irrigation Intervals on Maize Yield (zea mays L.) and some Soil Properties. J. Soil Sci. Agric. Eng. 11, 793–799. https://doi.org/10.21608/JSSAE.2020.160919

Eldor A. P., 2007. Soil Microbiology, Ecology, and Biochemistry, Elsevier.

Eldor, A.P., 2007. Soil Microbiology and Biochemistry.

Eriksson, T., Olsson, D., 2011. The Product Chains of Rare Earth Elements Used in Permanent Magnets and NiMH Batteries for Electric Vehicles.

Eslami, M., Khorassani, R., Coltorti, M., Malferrari, D., Faccini, B., Ferretti, G., Di Giuseppe, D., Fotovat, A., Halajnia, A., 2018. Leaching behaviour of a sandy soil amended with natural and NH4+ and K+ saturated clinoptilolite and chabazite. Arch. Agron. Soil Sci. https://doi.org/10.1080/03650340.2017.1414944

Esser, K. B.; Bockheim, J. G.; Helmke, P.A., 1991. ELEMENT DISTRIBUTION IN SOILS FORMED IN THE INDIANA DUNES, U.S.A. Soil Sci. Volume 152, 340–350.

Eyerusalem Goitom, 2011. Bioaccumulation of lanthanum in marbled crayfish (Procambarus sp.).

Fabani, M.P., Toro, M.E., Vázquez, F., Díaz, M.P., Wunderlin, D.A., 2009. Differential absorption of metals from soil to diverse vine varieties from the valley of tulum (Argentina): Consequences to evaluate wine provenance. J. Agric. Food Chem. 57, 7409–7416. https://doi.org/10.1021/JF901572K/ASSET/IMAGES/LARGE/JF-2009-01572K_0002.JPEG

Faccini, B., Di Giuseppe, D., Ferretti, G., Coltorti, M., Colombani, N., Mastrocicco, M., 2018. Natural and NH4+-enriched zeolitite amendment effects on nitrate leaching from a reclaimed agricultural soil (Ferrara Province, Italy). Nutr. Cycl. Agroecosystems 110, 327–341. https://doi.org/10.1007/s10705-017-9904-4

Farquhar, G.D., Ehleringer, J.R., Hubick, K.T., 1989. Carbon Isotope Discrimination and Photosynthesis. Annu. Rev. Plant Physiol. Plant Mol. Biol. 40, 503–537. https://doi.org/10.1146/annurev.pp.40.060189.002443

Farrés, M., Platikanov, S., Tsakovski, S., Tauler, R., 2015. Comparison of the variable importance in projection (VIP) and of the selectivity ratio (SR) methods for variable selection and interpretation. J. Chemom. 29, 528–536. https://doi.org/10.1002/CEM.2736

Fernández-Escobar, R., 2011. Use and abuse of nitrogen in olive fertilization. Acta Hortic. 888, 249–257. https://doi.org/10.17660/ActaHortic.2011.888.28

Fernández-Escobar, R., Beltrán, G., Sánchez-Zamora, M.A., García-Novelo, J., Aguilera, M.P., Uceda, M., 2006. Olive Oil Quality Decreases with Nitrogen Over-fertilization. HortScience 41, 215–219. https://doi.org/10.21273/HORTSCI.41.1.215

Fernández-Escobar, R., Benlloch, M., Herrera, E., García-Novelo, J.M., 2004. Effect of traditional and slow-release N fertilizers on growth of olive nursery plants and N losses by leaching. Sci. Hortic. (Amsterdam). 101, 39–49. https://doi.org/10.1016/j.scienta.2003.09.008

Fernández-Escobar, R., García-Novelo, J.M., Molina-Soria, C., Parra, M.A., 2012. An approach to nitrogen balance in olive orchards. Sci. Hortic. (Amsterdam). 135, 219–226. https://doi.org/10.1016/J.SCIENTA.2011.11.036

Ferreira, I.Q., Arrobas, M., Moutinho-Pereira, J.M., Correia, C.M., Rodrigues, M.Â., 2020. The effect of nitrogen applications on the growth of young olive trees and nitrogen use efficiency. Turkish J. Agric. For. 44, 278–289. https://doi.org/10.3906/tar-1905-26

Ferretti, G., Di Giuseppe, D., Faccini, B., Coltorti, M., 2018. Mitigation of sodium risk in a sandy agricultural soil by the use of natural zeolites. Environ. Monit. Assess. 190, 646. https://doi.org/10.1007/s10661-018-7027-2

Ferretti, G., Di Giuseppe, D., Natali, C., Faccini, B., Bianchini, G., Coltorti, M., 2017a. C-N elemental and isotopic investigation in agricultural soils: Insights on the effects of zeolitite amendments. Chemie der Erde 77, 45–52. https://doi.org/10.1016/j.chemer.2017.02.002

Ferretti, G., Faccini, B., Antisari, L.V., Di Giuseppe, D., Coltorti, M., 2019. 15N natural abundance, nitrogen and carbon pools in soil-sorghum system amended with natural and NH4+-enriched zeolitites. Appl. Sci. 9, 4524. https://doi.org/10.3390/app9214524

Ferretti, G., Galamini, G., Deltedesco, E., Gorfer, M., Fritz, J., Faccini, B., Mentler, A., Zechmeister-Boltenstern, S., Coltorti, M., Keiblinger, K.M., 2021. Gross Ammonification and Nitrification Rates in Soil Amended with Natural and NH4-Enriched Chabazite Zeolite and Nitrification Inhibitor DMPP. Appl. Sci. 11, 2605. https://doi.org/10.3390/app11062605

Ferretti, G., Keiblinger, K.M., Faccini, B., Di Giuseppe, D., Mentler, A., Zechmeister-Boltenstern, S., Coltorti, M., 2020. Effects of Different Chabazite Zeolite Amendments to Sorption of Nitrification Inhibitor 3,4-Dimethylpyrazole Phosphate (DMPP) in Soil. J. Soil Sci. Plant Nutr. https://doi.org/10.1007/s42729-020-00184-3

Ferretti, G., Keiblinger, K.M., Zimmermann, M., Di Giuseppe, D., Faccini, B., Colombani, N., Mentler, A., Zechmeister-Boltenstern, S., Coltorti, M., Mastrocicco, M., 2017b. High resolution short-term investigation of soil CO2, N2O, NOx and NH3 emissions after different chabazite zeolite amendments. Appl. Soil Ecol. 119, 138–144. https://doi.org/10.1016/j.apsoil.2017.06.004

Flexas, J., Hendrickson, L., Wah Soon Chow, 2001. Photoinactivation of photosystem II in high light-acclimated grapevines. Funct. Plant Biol. 28, 755–764. https://doi.org/10.1071/PP99210

Galamini, G., Ferretti, G., Medoro, V., Tescaro, N., Faccini, B., Coltorti, M., 2020. Isotherms, Kinetics, and Thermodynamics of NH4+ Adsorption in Raw Liquid Manure by Using Natural Chabazite Zeolite-Rich Tuff. Water 12, 2944. https://doi.org/10.3390/w12102944

Galgano, F., Favati, F., Caruso, M., Scarpa, T., Palma, A., 2008. Analysis of trace elements in southern Italian wines and their classification according to provenance. LWT - Food Sci. Technol. 41, 1808–1815. https://doi.org/10.1016/j.lwt.2008.01.015

Galli, E., Passaglia, E., 2011. Natural zeolites in environmental engineering, in: Harald Holzapfel (Ed.), Zeolites in Chemical Engineering. Verlag ProcessEng Engineering GmbH, pp. 392–416.

GeoViewer — Geoportale [WWW Document], n.d. URL https://geoportale.regione.emilia-romagna.it/mappe/geo-viewer?layer_id=0f536ddad2c442d293b6afa0119edde5 (accessed 8.16.22).

Giaccio, M., Vicentini, A., 2008. Determination of the geographical origin of wines by means of the mineral content and the stable isotope ratios: a review. J.Commodity Sci.Technol.Quality 47, 267–284.

Goldschmidt, V.M., 1937. The principles of distribution of chemical elements in minerals and rocks. The seventh Hugo Müller Lecture, delivered before the Chemical Society on March 17th, 1937. J. Chem. Soc. 655–673. https://doi.org/10.1039/JR9370000655

Gottardi, E. & Galli, G., 1985. Natural Zeolites. Minerals and Rocks Series Vol. 18. Geol. Mag. 123, 718–718. https://doi.org/10.1017/S0016756800024456

Gouvinhas, I., Domínguez-Perles, R., Machado, N., Carvalho, T., Matos, C., Barros, A.I.R.N.A., 2016. Effect of Agro-Environmental Factors on the Mineral Content of Olive Oils: Categorization of the Three Major Portuguese Cultivars. JAOCS, J. Am. Oil Chem. Soc. 93, 813–822. https://doi.org/10.1007/S11746-016-2827-4

Habran, A., Commisso, M., Helwi, P., Hilbert, G., Negri, S., Ollat, N., Gomès, E., van Leeuwen, C., Guzzo, F., Delrot, S., 2016. Roostocks/Scion/Nitrogen Interactions Affect Secondary Metabolism in the Grape Berry. Front. Plant Sci. 7. https://doi.org/10.3389/fpls.2016.01134

Hatfield, J.L., Follet, R.F., 2018. Nitrogen in the Environment, Angewandte Chemie International

Edition, 6(11), 951–952.

Haxel GB, Hedrick JB, O.G., 2002. Rare earth elements: critical resources for high technology.

Henderson, P., 1984. General Geochemical Properties and Abundances of the Rare Earth Elements. Rare Earth Elem. Geochemistry 2, 1–32. https://doi.org/10.1016/B978-0-444-42148-7.50006-X

Higueras, P., Amorós, J.A., Esbrí, J.M., García-Navarro, F.J., Pérez de los Reyes, C., Moreno, G., 2012. Time and space variations in mercury and other trace element contents in olive tree leaves from the Almadén Hg-mining district. J. Geochemical Explor. 123, 143–151. https://doi.org/10.1016/J.GEXPLO.2012.04.012

Hogberg, P., 1997. Tansley Review No. 95 15 N natural abundance in soil-plant systems. New Phytol. 137, 179–203. https://doi.org/10.1046/j.1469-8137.1997.00808.x

Hu, Z., Haneklaus, S., Sparovek, G., Schnug, E., 2006. Rare Earth Elements in Soils. https://doi.org/10.1080/00103620600628680 37, 1381–1420. https://doi.org/10.1080/00103620600628680

Hu, Z., Richter, H., Sparovek, G., Schnug, E., 2004. Physiological and Biochemical Effects of Rare Earth Elements on Plants and Their Agricultural Significance: A Review. J. Plant Nutr. 27, 183–220. https://doi.org/10.1081/PLN-120027555

Jackson, D.I., Lombard, P.B., Kabinett, L.Q., 1993. Environmental and management practices affecting grape composition and wine quality - A review. Am. J. Enol. Vitic. 44, 409–430.

Jagadeesha, B.G., Narayana, Y., Sudarshan, M., Banerjee, S., 2018. Studies on uptake and retention of trace elements by medicinal plants in the environs of Hassan of South India. Radiat. Phys. Chem. 144, 317–321. https://doi.org/10.1016/J.RADPHYSCHEM.2017.09.005

Jakkula V., W.S.P., 2018. Zeolites: Potential soil amendments for improving nutrient and water use efficiency and agriculture productivity [WWW Document]. Sci. Rev. Chem. Commun. URL https://www.researchgate.net/publication/325544295_Zeolites_Potential_soil_amendments_for_improving_nutrient_and_water_use_efficiency_and_agriculture_productivity (accessed 4.11.22).

Jakubowski, N., Brandt, R., Stuewer, D., Eschnauer, H.R., Görtges, S., 1999. Analysis of wines by ICP-MS: Is the pattern of the rare earth elements a reliable fingerprint for the provenance? Fresenius' J. Anal. Chem. 1999 3645 364, 424–428. https://doi.org/10.1007/S002160051361

Jalali-Heravi, M., Vosough, M., 2004. Characterization and determination of fatty acids in fish oil using gas chromatography–mass spectrometry coupled with chemometric resolution techniques. J. Chromatogr. A 1024, 165–176. https://doi.org/10.1016/J.CHROMA.2003.10.032

Janick, J., 1992. Horticultural reviews volume 14.

Kato, Y., Fujinaga, K., Nakamura, K., Takaya, Y., Kitamura, K., Ohta, J., Toda, R., Nakashima, T., Iwamori, H., 2011. Deep-sea mud in the Pacific Ocean as a potential resource for rare-earth elements. Nat. Geosci. 2011 48 4, 535–539. https://doi.org/10.1038/ngeo1185

Kelly, S., Heaton, K., Hoogewerff, J., 2005. Tracing the geographical origin of food: The application of multi-element and multi-isotope analysis. Trends Food Sci. Technol. 16, 555–567. https://doi.org/10.1016/J.TIFS.2005.08.008

Kesraoui-Ouki, S., Cheeseman, C.R., Perry, R., 1994. Natural zeolite utilisation in pollution control: A review of applications to metals' effluents. J. Chem. Technol. Biotechnol. https://doi.org/10.1002/jctb.280590202

Kizildeniz, T., Pascual, I., Irigoyen, J.J., Morales, F., 2021. Future <scp> CO_2 </scp> , warming and water deficit impact white and red Tempranillo grapevine: Photosynthetic acclimation to elevated <scp> CO_2 </scp> and biomass allocation. Physiol. Plant. 172, 1779–1794.

https://doi.org/10.1111/ppl.13388

Kliewer, W.M., Dokoozlian, N.K., 2005. Leaf area/crop weight ratios of grapevines: Influence on fruit composition and wine quality. Am. J. Enol. Vitic. 56, 170–181.

Kment, P., Mihaljevič, M., Ettler, V., Šebek, O., Strnad, L., Rohlová, L., 2005. Differentiation of Czech wines using multielement composition – A comparison with vineyard soil. Food Chem. 91, 157–165. https://doi.org/10.1016/J.FOODCHEM.2004.06.010

Kovaříková, M., Tomášková, I., Soudek, P., 2019. Rare earth elements in plants. https://doi.org/10.32615/bp.2019.003

Kulaksiz, S., Bau, M., 2011. Anthropogenic gadolinium as a microcontaminant in tap water used as drinking water in urban areas and megacities. Appl. Geochemistry 26, 1877–1885. https://doi.org/10.1016/J.APGEOCHEM.2011.06.011

Kumar, S., Singh, B., 2009. Effect of water stress on carbon isotope discrimination and Rubisco activity in bread and durum wheat genotypes. Physiol. Mol. Biol. Plants 2009 153 15, 281–286. https://doi.org/10.1007/S12298-009-0032-8

Lang, C.P., Merkt, N., Zörb, C., 2018. Different nitrogen (N) forms affect responses to N form and N supply of rootstocks and grafted grapevines. Plant Sci. 277, 311–321. https://doi.org/10.1016/J.PLANTSCI.2018.10.004

Lanza, B., Ninfali, P., 2020. Antioxidants in Extra Virgin Olive Oil and Table Olives: Connections between Agriculture and Processing for Health Choices. Antioxidants 9, 41. https://doi.org/10.3390/antiox9010041

Lavee, S., Wodner, M., 2015. Factors affecting the nature of oil accumulation in fruit of olive (Olea europaea L.) cultivars. http://dx.doi.org/10.1080/00221589.1991.11516187 66, 583–591. https://doi.org/10.1080/00221589.1991.11516187

Laveuf, C., Cornu, S., Guilherme, L.R.G., Guerin, A., Juillot, F., 2012. The impact of redox conditions on the rare earth element signature of redoximorphic features in a soil sequence developed from limestone. Geoderma 170, 25–38. https://doi.org/10.1016/J.GEODERMA.2011.10.014

Lechhab, T., Lechhab, W., Cacciola, F., Salmoun, F., 2022. Sets of internal and external factors influencing olive oil (Olea europaea L.) composition: a review. undefined 248, 1069–1088. https://doi.org/10.1007/S00217-021-03947-Z

Leita, L., Mondini, C., De Nobili, M., Simoni, A., Sequi, P., 1998. Heavy Metal Content in Xylem Sap (Vitis Vinifera) from Mining and Smelting Areas. Environ. Monit. Assess. 1998 502 50, 189–200. https://doi.org/10.1023/A:1005854213903

Lipin B. and McKay G., 1989. Geochemistry and Mineralogy of Rare Earth Elements. Mineralogical Society of America, Washington.

Llorent-Martínez, E.J., Ortega-Barrales, P., Fernández-De Córdova, M.L., Domínguez-Vidal, A., Ruiz-Medina, A., 2011. Investigation by ICP-MS of trace element levels in vegetable edible oils produced in Spain. Food Chem. 127, 1257–1262. https://doi.org/10.1016/j.foodchem.2011.01.064

Long, K.R., Van Gosen, B.S., Foley, N.K., Cordier, D., 2012. The principal rare earth elements deposits of the United States: A summary of domestic deposits and a global perspective. Non-Renewable Resour. Issues Geosci. Soc. Challenges 131–155. https://doi.org/10.1007/978-90-481-8679-2_7/COVER/

Lopes, J.I., Arrobas, M., Brito, C., Gonçalves, A., Silva, E., Martins, S., Raimundo, S., Rodrigues, M.Â., Correia, C.M., 2020. Mycorrhizal Fungi were More Effective than Zeolites in Increasing the Growth of Non-Irrigated Young Olive Trees. Sustain. 2020, Vol. 12, Page 10630 12, 10630. https://doi.org/10.3390/SU122410630

Luykx, D.M.A.M., van Ruth, S.M., 2008. An overview of analytical methods for determining the geographical origin of food products. Food Chem. 107, 897–911. https://doi.org/10.1016/J.FOODCHEM.2007.09.038

Ma, X., Jacoby, P.W., Sanguinet, K.A., 2020. Improving Net Photosynthetic Rate and Rooting Depth of Grapevines Through a Novel Irrigation Strategy in a Semi-Arid Climate. Front. Plant Sci. 11, 1–12. https://doi.org/10.3389/fpls.2020.575303

Mahesh, M., Thomas, J., Arun Kumar, K., Bhople, B.S., Saresh, N. V, Vaid, S.K., Sahu, S.K., 2018. Zeolite Farming: A Sustainable Agricultural Prospective. Int.J.Curr.Microbiol.App.Sci 7, 2912–2924. https://doi.org/10.20546/ijcmas.2018.705.340

Malferrari, D., Laurora, A., Brigatti, M.F., Coltorti, M., Di Giuseppe, D., Faccini, B., Passaglia, E., Vezzalini, M.G., 2013. Open-field experimentation of an innovative and integrated zeolitite cycle: Project definition and material characterization. Rend. Lincei 24, 141–150. https://doi.org/10.1007/s12210-013-0235-3

Mansour-Gueddes, S. Ben, Saidana-Naija, D., Flamini, G., Cheraief, I., Braham, M., 2022. Assessment of the climatic condition's impact on volatiles, polyphenols and mineral contents in tunisian olive tree (Olea europaea L.). Polish J. Environ. Stud. 31, 219–230. https://doi.org/10.15244/PJOES/133232

Martelli, L., Amorosi, A., Severi, P., Cazzoli, M.A., Colalongo, M.L., Vaiani, S.C., Alvisi, F., Bassetti, M., Fusco, F., 2009. NOTE ILLUSTRATIVE della CARTA GEOLOGICA D'ITALIA alla scala 1:50.000 foglio 221 BOLOGNA.

Masaaki Horikoshi and Yuan Tang, 2016. ggfortify: Data Visualization Tools for Statistical Analysis Results.

Mastrocicco, M., Colombani, N., Di Giuseppe, D., Faccini, B., Ferretti, G., Coltorti, M., 2016. Abnormal trace element concentrations in a shallow aquifer belonging to saline reclaimed environments, Codigoro (Italy). Rend. Lincei 27, 95–104. https://doi.org/10.1007/s12210-015-0454-x

Maxwell, T.M., Silva, L.C.R., Horwath, W.R., 2014. Using multielement isotopic analysis to decipher drought impacts and adaptive management in ancient agricultural systems. Proc. Natl. Acad. Sci. U. S. A. https://doi.org/10.1073/pnas.1417618111

Mazzi, F., Galli, E., 1983. THE TETRAHEDRAL FRAMEWORK OF CHABAZITE. TETRAHEDRAL Framew. CHABAZITE.

Mechri, B., Issaoui, M., Echbili, A., Chehab, H., Mariem, F.B., Braham, M., Hammami, M., 2009. Olive orchard amended with olive mill wastewater: Effects on olive fruit and olive oil quality. J. Hazard. Mater. 172, 1544–1550. https://doi.org/10.1016/J.JHAZMAT.2009.08.026

Medini, S., Janin, M., Verdoux, P., Techer, I., 2015. Methodological development for 87Sr/86Sr measurement in olive oil and preliminary discussion of its use for geographical traceability of PDO Nîmes (France). Food Chem. 171, 78–83. https://doi.org/10.1016/J.FOODCHEM.2014.08.121

Medoro, V., Ferretti, G., Galamini, G., Rotondi, A., Morrone, L., Faccini, B., Coltorti, M., 2022. Reducing Nitrogen Fertilization in Olive Growing by the Use of Natural Chabazite-Zeolitite as Soil Improver. L. 2022, Vol. 11, Page 1471 11, 1471.

Mercurio, M., Grilli, E., Odierna, P., Morra, V., Prohaska, T., Coppola, E., Grifa, C., Buondonno, A., Langella, A., 2014. A 'Geo-Pedo-Fingerprint' (GPF) as a tracer to detect univocal parent material-to-wine production chain in high quality vineyard districts, Campi Flegrei (Southern Italy). Geoderma 230–231, 64–78. https://doi.org/10.1016/J.GEODERMA.2014.04.006

Milosevic, T., Milosevic, N., 2009. The effect of zeolite, organic and inorganic fertilizers on soil chemical properties, growth and biomass yield of apple trees. Plant, Soil Environ. 55, 528–535. https://doi.org/10.17221/107/2009-PSE

Minardi, S., Haniati, I.L., Nastiti, A.H.L., 2020. Adding manure and zeolite to improve soil chemical properties and increase soybean yield. SainsTanah - J. Soil Sci. Agroclimatol. 17, 1. https://doi.org/10.20961/stjssa.v17i1.41087

Mir-Marqués, A., Elvira-Sáez, C., Cervera, M.L., Garrigues, S., de la Guardia, M., 2016. Authentication of protected designation of origin artichokes by spectroscopy methods. Food Control 59, 74–81. https://doi.org/10.1016/j.foodcont.2015.05.004

Morrone, L., Neri, L., Facini, O., Galamini, G., Medoro, V., 2021. Influence of zeolitite foliar coating on photosynthesis , VOC emission and quality of extra virgin olive oil.

Moshoeshoe, M., Silas Nadiye-Tabbiruka, M., Obuseng, V., 2017. A Review of the Chemistry, Structure, Properties and Applications of Zeolites. Am. J. Mater. Sci. 7, 196–221. https://doi.org/10.5923/j.materials.20170705.12

Mumpton, F.A., 1999. La roca magica: Uses of natural zeolites in agriculture and industry. Proc. Natl. Acad. Sci. U. S. A. 96, 3463–3470. https://doi.org/10.1073/pnas.96.7.3463

Mustafa, A., Hu, X., Shah, S.A.A., Abrar, M.M., Maitlo, A.A., Kubar, K.A., Saeed, Q., Kamran, M., Naveed, M., Boren, W., Nan, S., Minggang, X., 2021. Long-term fertilization alters chemical composition and stability of aggregate-associated organic carbon in a Chinese red soil: evidence from aggregate fractionation, C mineralization, and 13C NMR analyses. J. Soils Sediments 21, 2483–2496. https://doi.org/10.1007/S11368-021-02944-9/TABLES/3

Nasr, E.G., Epova, E.N., de Diego, A., Souissi, R., Hammami, M., Abderrazak, H., Donard, O.F.X., 2021. Trace elements analysis of tunisian and european extra virgin olive oils by icp-ms and chemometrics for geographical discrimination. Foods 11. https://doi.org/10.3390/foods11010082

Norman, A., Zou, X., Barnett, J., 2014. Critical minerals: rare earths and the U.S.

O'Leary, M.H., 1988. Carbon Isotopes in Photosynthesis. Bioscience 38, 328–336. https://doi.org/10.2307/1310735

Ogrinc, N., Košir, I.J., Spangenberg, J.E., Kidrič, J., 2003. The application of NMR and MS methods for detection of adulteration of wine, fruit juices, and olive oil. A review. Anal. Bioanal. Chem. 376, 424–430. https://doi.org/10.1007/S00216-003-1804-6/FIGURES/2

Othman, Y.A., Leskovar, D., 2019. Nitrogen management influenced root length intensity of young olive trees. Sci. Hortic. (Amsterdam). 246, 726–733. https://doi.org/10.1016/J.SCIENTA.2018.11.052

Ozaki, T., Enomoto, S., 2001. Uptake of rare earth elements by Dryopteris erythrosora (autumn fern). RIKEN Rev. No 35.

Passaglia, E., 2008. Zeoliti naturali, zeolititi e loro applicazioni. Arvan.

Patumi, M., D'Andria, R., Fontanazza, G., Morelli, G., Giorio, P., Sorrentino, G., 2015. Yield and oil quality of intensively trained trees of three cultivars of olive (Olea europaea L.) under different irrigation regimes. http://dx.doi.org/10.1080/14620316.1999.11511180 74, 729–737. https://doi.org/10.1080/14620316.1999.11511180

Patumi, M., D'Andria, R., Marsilio, V., Fontanazza, G., Morelli, G., Lanza, B., 2002. Olive and olive oil quality after intensive monocone olive growing (Olea europaea L., cv. Kalamata) in different irrigation regimes. Food Chem. 77, 27–34. https://doi.org/10.1016/S0308-8146(01)00317-X

Pavan, V., 2022. Rapporto IdroMeteoClima Emilia-Romagna. Dati 2021. Osservatorio clima di ARPAE.

Pavan, V., 2021. Rapporto IdroMeteoClima Emilia-Romagna. Dati 2020. Osservatorio clima di ARPAE.

Pavan, V., Marletto, V., 2020. Rapporto IdroMeteoClima Emilia-Romagna. Dati 2019. Osservatorio clima di ARPAE.

Peng, S., Buresh, R.J., Huang, J., Yang, J., Zou, Y., Zhong, X., Wang, G., Zhang, F., 2006. Strategies for overcoming low agronomic nitrogen use efficiency in irrigated rice systems in China. F. Crop. Res. 96, 37–47. https://doi.org/10.1016/j.fcr.2005.05.004

Pepi, S., Chicca, M., Piroddi, G., Tassinari, R., Vaccaro, C., 2019. Geographical origin of Vitis vinifera cv. Cannonau established by the index of bioaccumulation and translocation coefficients. Environ. Monit. Assess. 191. https://doi.org/10.1007/s10661-019-7544-7

Pepi, S., Coletta, A., Crupi, P., Leis, M., Russo, S., Sansone, L., Tassinari, R., Chicca, M., Vaccaro, C., 2016a. Geochemical characterization of elements in Vitis vinifera cv. Negroamaro grape berries grown under different soil managements. Environ. Monit. Assess. 188. https://doi.org/10.1007/s10661-016-5203-9

Pepi, S., Grisenti, P., Sansone, L., Chicca, M., Vaccaro, C., 2018. Chemical elements as fingerprints of geographical origin in cultivars of Vitis vinifera L. raised on the same SO4 rootstock. Environ. Sci. Pollut. Res. 25, 490–506. https://doi.org/10.1007/s11356-017-0443-y

Pepi, S., Sansone, L., Chicca, M., Marrocchino, E., Vaccaro, C., 2016b. Distribution of rare earth elements in soil and grape berries of Vitis vinifera cv. "Glera". Environ. Monit. Assess. 188. https://doi.org/10.1007/s10661-016-5490-1

Pepi, S., Sansone, L., Chicca, M., Vaccaro, C., 2017. Relationship among geochemical elements in soil and grapes as terroir fingerprintings in Vitis vinifera L. cv. "Glera". Chemie der Erde 77, 121–130. https://doi.org/10.1016/j.chemer.2017.01.003

Perez-Caballero, R., Gil, J., Benitez, C., Gonzalez, J.L., 2008. The effect of adding zeolite to soils in order to improve the N-K nutrition of olive trees. Preliminary results. Am. J. Agric. Biol. Sci. 3, 321–324. https://doi.org/10.3844/ajabssp.2008.321.324

Poblete-Echeverría, C., Espinace, D., Sepúlveda-Reyes, D., Zúñiga, M., Sanchez, M., 2017. Analysis of crop water stress index (CWSI) for estimating stem water potential in grapevines: comparison between natural reference and baseline approaches. Acta Hortic. 189–194. https://doi.org/10.17660/ActaHortic.2017.1150.27

Polat, E., Karaca, M., Demir, H., and O., A.N., 2004. Use of Natural Zeolite (Clinoptilolite) in Agriculture. J. fruit Ornam. plant Res. 12, 183–189.

Pošćić, F., Žanetić, M., Fiket, Ž., Furdek Turk, M., Mikac, N., Bačić, N., Lučić, M., Romić, M., Bakić, H., Jukić Špika, M., Urlić, B., Runjić, M., Vuletin Selak, G., Vitanović, E., Klepo, T., Rošin, J., Rengel, Z., Perica, S., 2020. Accumulation and partitioning of rare earth elements in olive trees and extra virgin olive oil from Adriatic coastal region. Plant Soil 448, 133–151. https://doi.org/10.1007/s11104-019-04418-x

Prentki, M., Madiraju, M.S.R., 2012. Glycerolipid/free fatty acid cycle and islet β-cell function in health, obesity and diabetes. Mol. Cell. Endocrinol. 353, 88–100. https://doi.org/10.1016/J.MCE.2011.11.004

Prisa, D., 2020. Optimised fertilisation with zeolitites containing Plant Growth Promoting Rhizobacteria (PGPR) in Ranunculus asiaticus. https://gsconlinepress.com/journals/gscbps/sites/default/files/GSCBPS-2020-0011.pdf 10, 096–102. https://doi.org/10.30574/GSCBPS.2020.10.1.0011

Protano, G., Rossi, S., 2014. Relationship between soil geochemistry and grape composition in Tuscany (Italy). J. Plant Nutr. Soil Sci. 177, 500–508. https://doi.org/10.1002/JPLN.201300055

Punturo, R., D'Antone, C., Pepi, S., Vaccaro, C., 2018. Rare earth elements absorption patterns in grapevine "Vitis vinifera L." cultivated in carbonate terrains (south-eastern Sicily, Italy). Environ. Earth Sci. 77. https://doi.org/10.1007/s12665-018-7989-y

Regattieri, A., Gamberi, M., Manzini, R., 2007. Traceability of food products: General framework and experimental evidence. J. Food Eng. 81, 347–356. https://doi.org/10.1016/J.JFOODENG.2006.10.032

Reháková, M., Čuvanová, S., Dzivák, M., Rimár, J., Gaval'Ová, Z., 2004. Agricultural and agrochemical uses of natural zeolite of the clinoptilolite type. Curr. Opin. Solid State Mater. Sci. 8, 397–404. https://doi.org/10.1016/j.cossms.2005.04.004

Riehl, S., Pustovoytov, K.E., Weippert, H., Klett, S., Hole, F., 2014. Drought stress variability in ancient Near Eastern agricultural systems evidenced by δ13C in barley grain. Proc. Natl. Acad. Sci. U. S. A. 111, 12348–12353. https://doi.org/10.1073/pnas.1409516111

Robinson, W.O., Bastron, H., Murata, K.J., 1958. Biogeochemistry of the rare-earth elements with particular reference to hickory trees. Geochim. Cosmochim. Acta 14, 55–67. https://doi.org/10.1016/0016-7037(58)90093-0

Robinson, W.O., Whetstone, R., Scribner, B.F., 1938. The Presence of Rare Earths in Hickory Leaves. Science (80-.). 87, 470–470. https://doi.org/10.1126/SCIENCE.87.2264.470.A

Rodrigues, M.Â., Torres, L. do N.D., Damo, L., Raimundo, S., Sartor, L., Cassol, L.C., Arrobas, M., 2021. Nitrogen Use Efficiency and Crop Yield in Four Successive Crops Following Application of Biochar and Zeolites. J. Soil Sci. Plant Nutr. 21, 1053–1065. https://doi.org/10.1007/S42729-021-00421-3/FIGURES/6

Rodrigues, S.M., Otero, M., Alves, A.A., Coimbra, J., Coimbra, M.A., Pereira, E., Duarte, A.C., 2011. Elemental analysis for categorization of wines and authentication of their certified brand of origin. J. Food Compos. Anal. 24, 548–562. https://doi.org/10.1016/J.JFCA.2010.12.003

Rosati, A., Cafiero, C., Paoletti, A., Alfei, B., Caporali, S., Casciani, L., Valentini, M., 2014. Effect of agronomical practices on carpology, fruit and oil composition, and oil sensory properties, in olive (Olea europaea L.). Food Chem. 159, 236–243. https://doi.org/10.1016/J.FOODCHEM.2014.03.014

Rotondi, A., Morrone, L., Facini, O., Faccini, B., Ferretti, G., Coltorti, M., 2021. Distinct particle films impacts on olive leaf optical properties and plant physiology. Foods. https://doi.org/10.3390/foods10061291

Ruiz-Perez, D., Guan, H., Madhivanan, P., Mathee, K., Narasimhan, G., 2020. So you think you can PLS-DA? BMC Bioinformatics 21, 1–10. https://doi.org/10.1186/S12859-019-3310-7/FIGURES/6

S., F., Trujillo, J.P., E., P., Conde, J., 2001. Classification and differentiation of bottled sweet wines of Canary Islands (Spain) by their metallic content. Eur. Food Res. Technol. 213, 145–149. https://doi.org/10.1007/s002170100344

Sacco, D., Brescia, M.A., Sgaramella, A., Casiello, G., Buccolieri, A., Ogrinc, N., Sacco, A., 2009. Discrimination between Southern Italy and foreign milk samples using spectroscopic and analytical data. Food Chem. 114, 1559–1563. https://doi.org/10.1016/J.FOODCHEM.2008.11.056

Samman, S., Chow, J.W.Y., Foster, M.J., Ahmad, Z.I., Phuyal, J.L., Petocz, P., 2008. Fatty acid composition of edible oils derived from certified organic and conventional agricultural methods. Food Chem. 109, 670–674. https://doi.org/10.1016/J.FOODCHEM.2007.12.067

Semhi, K., Clauer, N., Chaudhuri, S., 2012. Variable element transfers from an illite-rich substrate to growing plants during a three-month experiment. Appl. Clay Sci. 57, 17–24. https://doi.org/10.1016/J.CLAY.2011.12.002

Sepaskhah, A.R., Barzegar, M., 2010. Yield, water and nitrogen-use response of rice to zeolite and nitrogen fertilization in a semi-arid environment. Agric. Water Manag. 98, 38–44. https://doi.org/10.1016/J.AGWAT.2010.07.013

Shan, Y., Huang, M., Suo, L., Zhao, X., Wu, L., 2019. Composition and variation of soil δ15N stable isotope in natural ecosystems. CATENA 183, 104236. https://doi.org/10.1016/J.CATENA.2019.104236

Sharma, L.K., Bali, S.K., 2018. A review of methods to improve nitrogen use efficiency in agriculture. Sustain. 10. https://doi.org/10.3390/SU10010051

Shi, L., Liu, L., Yang, B., Sheng, G., Xu, T., 2020. Evaluation of Industrial Urea Energy Consumption (EC) Based on Life Cycle Assessment (LCA). Sustainability 12, 3793. https://doi.org/10.3390/su12093793

Soares, J.R., Cantarella, H., Menegale, M.L. de C., 2012. Ammonia volatilization losses from surface-applied urea with urease and nitrification inhibitors. Soil Biol. Biochem. 52, 82–89. https://doi.org/10.1016/j.soilbio.2012.04.019

Soto-Vaca, A., Losso, J.N., McDonough, K., Finley, J.W., 2013. Differential effect of 14 free fatty acids in the expression of inflammation markers on human arterial coronary cells. J. Agric. Food Chem. 61, 10074–10079. https://doi.org/10.1021/JF402966R/ASSET/IMAGES/LARGE/JF-2013-02966R_0005.JPEG

Szpak, P., 2014. Complexities of nitrogen isotope biogeochemistry in plant-soil systems: implications for the study of ancient agricultural and animal management practices. Front. Plant Sci. 5, 1–19. https://doi.org/10.3389/fpls.2014.00288

Taiyun Wei and Viliam Simko, 2021. R package ‘corrplot’: Visualization of a Correlation Matrix (Version 0.90).

Tarocco, A. and Aprea, A., 2021. Carta dei suoli della regione emilia-romagna.

Taylor, V.F., Longerich, H.P., Greenough, J.D., 2003. Multielement analysis of Canadian wines by inductively coupled plasma mass spectrometry (ICP-MS) and multivariate statistics. J. Agric. Food Chem. 51, 856–860. https://doi.org/10.1021/JF025761V/ASSET/IMAGES/LARGE/JF025761VF00003.JPEG

Thiel, G., Geisler, G., Blechschmidt, I., Danzer, K., 2004. Determination of trace elements in wines and classification according to their provenance. Anal. Bioanal. Chem. 378, 1630–1636. https://doi.org/10.1007/S00216-003-2444-6

Turra, C., 2018. Sustainability of rare earth elements chain: from production to food–a review. Int. J. Environ. Health Res. 28, 23–42. https://doi.org/10.1080/09603123.2017.1415307

Turra, C., 2017. Sustainability of rare earth elements chain: from production to food – a review. https://doi.org/10.1080/09603123.2017.1415307 28, 23–42. https://doi.org/10.1080/09603123.2017.1415307

Turra, C., de Nadai Fernandes, E.A., Bacchi, M.A., Sarriés, G.A., Júnior, F.B., Reyes, A.E.L., 2013. RARE EARTH ELEMENTS IN CITRUS PRODUCTION SYSTEMS. http://dx.doi.org/10.1080/01904167.2012.754040 36, 762–771. https://doi.org/10.1080/01904167.2012.754040

Turra, C., Fernandes, E.A.N., Bacchi, M.A., 2011. Evaluation on rare earth elements of Brazilian agricultural supplies. J. Environ. Chem. Ecotoxicol. 3, 86–92.

Tyler, G., 2004. Rare earth elements in soil and plant systems - A review, Plant and Soil.

Vincent Q. Vu, 2011. ggbiplot: A ggplot2 based biplot.

Vrignon-Brenas, S., Fontez, B., Bisson, A., Rolland, G., Chopard, J., Fumey, D., Metay, A., Pellegrino, A., 2022. Quantification of the pluriannual dynamics of grapevine growth responses to nitrogen supply using a Bayesian approach. J. Exp. Bot. 73, 1385–1401. https://doi.org/10.1093/jxb/erab469

Vrignon-Brenas, S., Metay, A., Leporatti, R., Gharibi, S., Fraga, A., Dauzat, M., Rolland, G., Pellegrino, A., 2019. Gradual responses of grapevine yield components and carbon status to nitrogen supply. Oeno One 53, 289–306. https://doi.org/10.20870/oeno-one.2019.53.2.2431

Vystavna, Y., Rätsep, R., Klymenko, N., Drozd, O., Pidlisnyuk, V., Klymenko, M., 2015. Comparison of soil-to-root transfer and translocation coefficients of trace elements in vines of Chardonnay and Muscat white grown in the same vineyard. Sci. Hortic. (Amsterdam). 192, 89–96. https://doi.org/10.1016/J.SCIENTA.2015.05.019

Wabaidur, S.M., AlAmmari, A., Aqel, A., AL-Tamrah, S.A., Alothman, Z.A., Ahmed, A.Y.B.H., 2016. Determination of free fatty acids in olive oils by UPHLC–MS. J. Chromatogr. B 1031, 109–115. https://doi.org/10.1016/J.JCHROMB.2016.07.040

Wang, L., Liang, T., 2015. Geochemical fractions of rare earth elements in soil around a mine tailing in Baotou, China. Sci. Reports 2015 51 5, 1–11. https://doi.org/10.1038/srep12483

Wang, Y., Zhang, M., Wang, X., 2000. Population growth responses of Tetrahymena shanghaiensis in exposure to rare earth elements. Biol. Trace Elem. Res. 2000 751 75, 265–275. https://doi.org/10.1385/BTER:75:1-3:265

Wang, Z., Zhang, X., Mu, Y., 2008. Effects of rare-earth fertilizers on the emission of nitrous oxide from agricultural soils in China. Atmos. Environ. 42, 3882–3887. https://doi.org/10.1016/J.ATMOSENV.2008.01.018

Wermelinger, B., Baumgärtner, J., Gutierrez, A.P., 1991. A demographic model of assimilation and allocation of carbon and nitrogen in grapevines. Ecol. Modell. 53, 1–26. https://doi.org/10.1016/0304-3800(91)90138-Q

Wickham, H., 2016. ggplot2: Elegant Graphics for Data Analysis.

World Reference Base | FAO SOILS PORTAL | Food and Agriculture Organization of the United Nations [WWW Document], n.d. URL https://www.fao.org/soils-portal/data-hub/soil-classification/world-reference-base/en/ (accessed 8.16.22).

Xinde, C., Xiaorong, W., Guiwen, Z., 2000. Assessment of the bioavailability of rare earth elements in soils by chemical fractionation and multiple regression analysis. Chemosphere 40, 23–28. https://doi.org/10.1016/S0045-6535(99)00225-8

Yang, J., Liu, Q., Zhang, L., Wu, S., Qi, M., Lu, S., Xi, Q., Cai, Y., 2009. Lanthanum chloride impairs memory, decreases pCaMK IV, pMAPK and pCREB expression of hippocampus in rats. Toxicol. Lett. 190, 208–214. https://doi.org/10.1016/J.TOXLET.2009.07.016

Zheng, J., Chen, T., Wu, Q., Yu, J., Chen, W., Chen, Y., Siddique, K.H.M., Meng, W., Chi, D., Xia, G., 2018. Effect of zeolite application on phenology, grain yield and grain quality in rice under water stress. Agric. Water Manag. 206, 241–251. https://doi.org/10.1016/J.AGWAT.2018.05.008

Zhou, B., Li, Z., Chen, C., 2017. Global potential of rare earth resources and rare earth demand from clean technologies. Minerals 7. https://doi.org/10.3390/MIN7110203

Treatment	Area	Type	Year	Management	pH	st.dev	EC (mS)	st.dev	SOM	st.dev	SOC	st.dev	TC (%)	st.dev	TN (%)	st.dev	$\delta^{13}C$	st.dev	$\delta^{15}N$	st.dev
cnt	SL	soil	2019	Pre-Fert	8.07	0.46	0.85	0.14	4.38	0.55	2.54	0.32	2.180	1.280	0.177	0.074	-20.320	4.125	6.947	1.933
zeo	SL	soil	2019	Pre-Fert	8.19	0.29	0.80	0.12	4.38	0.61	2.54	0.35	1.793	0.101	0.147	0.055	-16.733	4.546	4.960	0.653
cnt	SL	soil	2019	Post-Fert	7.42	0.73	2.12	1.74	5.97	1.14	3.46	0.66	2.543	0.190	0.207	0.049	-21.410	3.160	5.460	0.468
zeo	SL	soil	2019	Post-Fert	8.19	0.29	0.80	0.12	0.12	0.61	3.12	0.49	1.710	0.769	0.217	0.125	-20.017	3.040	5.673	0.499
cnt	SL	soil	2019	Harvest	7.78	0.49	0.89	0.47	2.96	0.51	1.71	0.30	1.290	0.573	0.103	0.025	-18.857	6.656	6.057	0.371
zeo	SL	soil	2019	Harvest	8.11	0.36	0.71	0.14	2.83	0.67	1.64	0.39	1.277	0.457	0.087	0.025	-15.010	7.553	4.747	0.176
cnt	SL	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid-19	covid-19
zeo	SL	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid-19	covid-19
cnt	SL	soil	2020	Post-Fert	8.21	0.50	9.14	5.34	4.47	0.21	2.59	0.12	1.417	0.381	0.289	0.148	-22.350	0.760	13.703	9.328
zeo	SL	soil	2020	Post-Fert	8.85	0.13	7.11	2.22	4.10	0.69	2.38	0.40	1.277	0.212	0.326	0.032	-22.190	0.389	17.577	3.932
cnt	SL	soil	2020	Harvest	7.60	0.40	3.41	2.83	2.77	0.70	1.60	0.41	0.939	0.135	0.157	0.033	-23.746	1.232	8.213	3.508
zeo	SL	soil	2020	Harvest	7.26	0.08	4.31	0.82	2.63	0.06	1.53	0.03	1.014	0.392	0.180	0.055	-25.887	1.145	9.417	4.206
cnt	SL	soil	2021	Pre-Fert	7.94	0.26	1.03	0.29	2.80	0.35	1.62	0.20	1.297	0.187	0.147	0.021	-22.871	0.759	5.358	0.384
zeo	SL	soil	2021	Pre-Fert	7.67	0.60	1.19	0.32	3.70	0.20	2.15	0.12	1.362	0.048	0.175	0.025	-25.008	3.243	7.311	1.956
cnt	SL	soil	2021	Post-Fert	7.50	0.25	0.64	0.16	4.71	1.06	2.73	0.62	1.442	0.371	0.172	0.019	-23.486	0.824	3.418	0.770
zeo	SL	soil	2021	Post-Fert	7.76	0.99	0.61	0.13	5.32	0.36	3.08	0.21	1.083	0.202	0.140	0.007	-22.457	3.358	2.953	1.083
cnt	SL	soil	2021	Harvest	7.30	0.20	0.03	0.02	3.92	0.56	2.27	0.33	0.762	0.207	0.109	0.022	-21.797	0.916	-0.317	2.695
zeo	SL	soil	2021	Harvest	7.29	0.10	0.05	0.02	3.60	0.43	2.09	0.25	0.847	0.289	0.117	0.005	-22.567	1.773	-4.020	4.691
cnt	BG	soil	2019	Pre-Fert	8.91	0.15	1.11	0.13	5.53	0.40	3.20	0.23	3.495	0.233	0.095	0.007	-4.265	0.361	2.895	0.304
zeo	BG	soil	2019	Pre-Fert	8.72	0.30	1.04	0.07	5.90	0.71	3.42	0.41	3.610	0.325	0.120	0.028	-5.915	1.690	4.225	1.435
cnt	BG	soil	2019	Post-Fert	8.82	0.20	1.40	0.24	5.01	0.59	2.91	0.34	3.603	0.255	0.108	0.028	-4.910	3.182	2.680	1.817
zeo	BG	soil	2019	Post-Fert	8.76	0.15	1.38	0.29	5.05	0.66	2.93	0.38	3.550	0.290	0.123	0.014	-5.880	1.930	2.670	1.994
cnt	BG	soil	2019	Harvest	8.83	0.06	1.36	0.66	6.30	0.93	3.65	0.54	3.800	0.014	0.130	0.014	-6.290	1.174	2.895	0.177
zeo	BG	soil	2019	Harvest	8.89	0.10	1.14	0.17	6.30	0.62	3.65	0.36	3.720	0.608	0.125	0.021	-6.130	1.697	3.290	2.899
cnt	BG	soil	2020	Pre-Fert	9.08	0.07	0.85	0.11	5.53	0.54	3.20	0.31	4.000	0.000	0.139	0.000	-8.260	0.000	6.040	0.000
zeo	BG	soil	2020	Pre-Fert	9.01	0.09	0.90	0.10	7.33	0.90	4.25	0.52	4.820	0.000	0.172	0.000	-11.760	0.000	5.450	0.000
cnt	BG	soil	2020	Post-Fert	8.45	0.19	2.44	0.21	5.60	0.26	3.25	0.15	4.003	0.262	0.143	0.018	-6.993	2.310	3.097	0.244
zeo	BG	soil	2020	Post-Fert	8.61	0.32	1.67	0.59	6.23	0.45	3.61	0.26	3.987	0.309	0.140	0.029	-7.490	2.327	1.693	2.681
cnt	BG	soil	2020	Harvest	8.75	0.22	1.36	0.28	5.33	1.44	3.09	0.83	4.062	0.537	0.125	0.026	-7.370	3.219	3.152	0.514
zeo	BG	soil	2020	Harvest	8.70	0.29	1.36	0.24	5.87	1.16	3.40	0.67	4.158	0.088	0.148	0.013	-8.524	0.799	4.471	1.061
cnt	BG	soil	2021	Pre-Fert	8.51	0.35	1.20	0.09	3.23	1.07	1.88	0.62	4.702	0.296	0.164	0.033	-6.729	1.994	5.190	0.782
zeo	BG	soil	2021	Pre-Fert	8.59	0.15	1.20	0.18	3.30	0.96	1.91	0.56	5.193	0.426	0.188	0.019	-8.340	1.758	5.413	0.349
cnt	BG	soil	2021	Post-Fert	8.70	0.08	1.13	0.29	5.17	2.33	3.00	1.35	4.582	0.386	0.168	0.008	-9.314	0.935	3.605	1.687
zeo	BG	soil	2021	Post-Fert	8.64	0.25	1.80	1.06	4.37	0.70	2.54	0.40	3.937	0.212	0.155	0.025	-7.549	1.516	3.615	1.817
cnt	BG	soil	2021	Harvest	8.74	0.45	0.08	0.02	6.10	0.53	3.54	0.31	3.697	0.163	0.117	0.016	-6.759	1.695	3.332	0.537
zeo	BG	soil	2021	Harvest	8.95	0.15	0.07	0.01	6.30	1.83	3.65	1.06	3.727	0.326	0.146	0.013	-9.058	1.386	9.577	3.498
cnt	BN	soil	2019	Pre-Fert	9.69	0.16	1.21	0.30	6.42	0.81	3.72	0.47	4.393	1.859	0.227	0.064	-13.160	3.820	2.660	0.959
zeo	BN	soil	2019	Pre-Fert	9.73	0.21	1.23	0.14	6.53	0.49	3.79	0.29	3.780	1.373	0.197	0.025	-11.400	2.455	2.207	0.426

A

cnt	BN	soil	2019	Post-Fert	9.71	0.17	1.18	0.23	7.13	0.59	4.14	0.34	4.073	1.927	0.167	0.006	-11.420	4.661	3.340	0.139
zeo	BN	soil	2019	Post-Fert	9.55	0.17	1.73	0.76	8.37	3.17	4.85	1.84	3.713	1.253	0.160	0.010	-11.700	2.998	3.607	0.888
cnt	BN	soil	2019	Harvest	9.74	0.20	1.08	0.12	6.43	0.81	3.73	0.47	4.010	1.589	0.160	0.000	-10.687	3.972	12.560	0.404
zeo	BN	soil	2019	Harvest	9.67	0.22	1.11	0.12	6.18	0.75	3.58	0.44	3.800	1.220	0.160	0.010	-11.630	2.291	11.607	6.872
cnt	BN	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid-19	covid-19
zeo	BN	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid-	covid	covid	covid-19	covid-19
cnt	BN	soil	2020	Post-Fert	8.11	0.14	0.33	0.18	7.23	0.70	4.20	0.41	4.260	1.895	0.264	0.086	-15.703	6.685	3.767	2.205
zeo	BN	soil	2020	Post-Fert	7.97	0.28	0.41	0.05	7.27	0.64	4.21	0.37	3.663	1.660	0.289	0.061	-14.483	4.186	2.593	1.635
cnt	BN	soil	2020	Harvest	7.98	0.09	0.13	0.06	2.63	1.89	1.53	1.09	4.512	2.021	0.199	0.019	-19.926	8.932	2.820	0.899
zeo	BN	soil	2020	Harvest	7.98	0.28	0.27	0.15	1.00	1.00	0.58	0.58	3.771	1.512	0.207	0.042	-12.833	4.018	3.734	2.078
cnt	BN	soil	2021	Pre-Fert	7.98	0.09	0.13	0.06	6.47	0.95	3.75	0.55	4.512	2.021	0.199	0.019	-19.926	8.932	2.820	0.899
zeo	BN	soil	2021	Pre-Fert	7.98	0.28	0.27	0.15	6.07	0.47	3.52	0.27	3.771	1.512	0.207	0.042	-12.833	4.018	3.734	2.078
cnt	BN	soil	2021	Post-Fert	8.13	0.23	0.37	0.20	6.53	0.31	3.79	0.18	4.572	1.997	0.246	0.064	-12.707	5.639	3.085	3.104
zeo	BN	soil	2021	Post-Fert	7.83	0.15	0.74	0.27	7.29	0.72	4.23	0.42	4.438	1.491	0.351	0.086	-15.543	3.725	4.696	3.195
cnt	BN	soil	2021	Harvest	7.70	0.27	0.07	0.04	6.99	0.52	4.06	0.30	4.080	2.150	0.222	0.054	-23.450	11.793	0.157	4.560
zeo	BN	soil	2021	Harvest	7.64	0.21	0.10	0.05	7.19	0.80	4.17	0.46	3.537	1.656	0.232	0.070	-21.487	10.293	2.120	2.530

B

Treatment	Area	Type	Year	Management	F^-	st.dev	Cl^-	st.dev	$N\text{-}NO_2$	st.dev	Br^-	st.dev	$N\text{-}NO_3$	st.dev	PO_4^{3-}	st.dev	SO_4^{2-}	st.dev
cnt	SL	soil	2019	Pre-Fert	6.07	1.39	14.06	14.98	0.37	0.09	0.07	0.15	17.20	11.69	11.21	17.78	20.36	6.81
zeo	SL	soil	2019	Pre-Fert	6.13	1.96	16.16	8.18	0.39	0.11	0.00	0.00	12.61	8.12	3.55	3.11	20.43	13.27
cnt	SL	soil	2019	Post-Fert	5.32	2.42	11.30	3.40	0.29	0.23	0.10	0.18	78.55	88.47	299.24	353.71	108.08	74.00
zeo	SL	soil	2019	Post-Fert	4.11	1.09	10.80	7.74	0.31	0.27	0.03	0.09	47.71	44.04	271.12	390.73	76.01	91.36
cnt	SL	soil	2019	Harvest	6.45	2.11	6.99	1.16	0.40	0.27	0.00	0.00	2.21	1.71	88.92	105.58	76.27	127.62
zeo	SL	soil	2019	Harvest	7.22	2.30	8.59	3.03	0.26	0.17	0.00	0.00	2.57	2.54	14.43	6.41	27.58	18.60
cnt	SL	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid
zeo	SL	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid
cnt	SL	soil	2020	Post-Fert	4.71	3.48	52.74	11.15	2.87	4.05	0.00	0.00	70.30	61.29	458.01	443.15	1781.88	982.12
zeo	SL	soil	2020	Post-Fert	4.76	0.51	26.05	9.24	1.92	1.38	0.00	0.00	46.76	1.58	203.63	98.18	897.00	532.02
cnt	SL	soil	2020	Harvest	2.17	1.07	22.90	7.37	2.25	1.55	0.00	0.00	205.39	182.09	10.41	11.09	592.65	456.14
zeo	SL	soil	2020	Harvest	2.32	0.41	20.17	8.70	2.30	0.18	0.00	0.00	399.98	86.84	12.62	17.41	102.78	52.71
cnt	SL	soil	2021	Pre-Fert	3.78	2.19	22.88	16.84	0.06	0.10	0.00	0.00	11.95	14.14	48.05	38.76	34.62	17.54
zeo	SL	soil	2021	Pre-Fert	2.18	0.84	14.19	6.81	0.76	0.27	0.00	0.00	18.88	11.73	123.44	133.33	99.89	91.95
cnt	SL	soil	2021	Post-Fert	3.50	1.12	15.55	4.51	1.34	0.10	0.24	0.42	19.36	6.45	49.42	18.25	55.59	33.91
zeo	SL	soil	2021	Post-Fert	4.69	2.47	16.16	5.29	3.19	2.99	8.78	15.20	22.32	9.09	35.69	35.54	37.00	29.07
cnt	SL	soil	2021	Harvest	4.20	1.29	22.14	3.57	0.56	0.60	na	na	5.10	1.83	22.46	25.06	23.28	12.40
zeo	SL	soil	2021	Harvest	2.35	0.65	39.77	24.01	0.17	0.00	na	na	7.65	4.45	15.48	10.11	17.57	14.64
cnt	BG	soil	2019	Pre-Fert	7.46	2.23	14.41	5.68	0.67	0.42	1.07	1.51	2.19	0.96	3.73	3.96	14.14	8.47
zeo	BG	soil	2019	Pre-Fert	6.18	1.70	12.66	6.06	0.56	0.07	0.25	0.09	3.14	1.31	2.39	2.07	12.05	0.98
cnt	BG	soil	2019	Post-Fert	6.11	1.78	44.40	13.97	0.44	0.40	1.11	0.03	14.78	12.47	4.72	3.82	59.82	27.72
zeo	BG	soil	2019	Post-Fert	5.16	1.80	37.39	10.91	0.55	0.19	0.17	0.07	15.76	10.41	5.10	5.72	90.58	52.23
cnt	BG	soil	2019	Harvest	7.38	2.55	35.64	3.40	0.00	0.00	0.12	0.06	3.46	3.30	0.80	0.95	40.20	22.51
zeo	BG	soil	2019	Harvest	5.72	1.99	54.61	12.18	0.17	0.33	0.29	0.36	3.64	3.06	1.11	2.22	61.32	30.30
cnt	BG	soil	2020	Pre-Fert	na	na	na	na	na	na	na	na	na	na	na	na	na	na
zeo	BG	soil	2020	Pre-Fert	na	na	na	na	na	na	na	na	na	na	na	na	na	na
cnt	BG	soil	2020	Post-Fert	3.06	0.88	20.96	4.11	0.23	0.39	0.00	0.00	105.00	22.48	0.10	0.17	34.70	9.68
zeo	BG	soil	2020	Post-Fert	4.79	0.74	24.00	11.09	1.85	3.20	0.00	0.00	38.62	40.92	0.00	0.00	27.01	2.22
cnt	BG	soil	2020	Harvest	4.61	0.98	33.28	4.73	0.43	0.05	0.00	0.00	20.32	9.73	1.90	1.95	35.09	16.21
zeo	BG	soil	2020	Harvest	3.97	0.72	27.57	11.98	0.00	0.00	0.00	0.00	42.35	34.39	2.40	4.16	24.31	10.85

C

cnt	BG	soil	2021	Pre-Fert	6.14	3.02	34.92	2.31	0.46	0.41	0.00	0.00	11.16	8.56	2.21	2.00	14.28	10.21
zeo	BG	soil	2021	Pre-Fert	4.96	0.97	18.79	10.69	0.00	0.00	0.00	0.00	7.68	4.81	5.58	1.49	14.43	4.71
cnt	BG	soil	2021	Post-Fert	4.29	1.13	18.91	4.64	2.15	0.76	0.00	0.00	24.95	15.38	2.74	3.16	50.44	36.09
zeo	BG	soil	2021	Post-Fert	4.67	1.43	21.82	3.84	2.62	1.59	0.00	0.00	102.34	88.46	0.00	0.00	43.24	34.68
cnt	BG	soil	2021	Harvest	2.39	1.01	54.90	38.21	0.00	0.00	0.14	0.00	4.91	4.05	5.67	0.00	19.49	5.73
zeo	BG	soil	2021	Harvest	3.04	0.81	51.08	44.57	0.30	0.00	0.00	0.00	12.08	7.96	6.17	5.01	27.32	7.92
cnt	BN	soil	2019	Pre-Fert	6.67	1.27	15.45	12.84	1.26	0.99	0.00	0.00	6.64	6.23	0.08	0.12	5.17	2.76
zeo	BN	soil	2019	Pre-Fert	6.43	1.15	12.70	6.55	0.76	0.31	0.00	0.00	8.41	7.57	0.05	0.07	3.86	0.67
cnt	BN	soil	2019	Post-Fert	5.69	1.39	9.78	4.53	0.42	0.14	0.00	0.00	29.07	22.05	0.03	0.10	3.34	1.15
zeo	BN	soil	2019	Post-Fert	5.81	1.49	13.43	7.53	0.51	0.21	0.00	0.00	57.82	69.69	0.05	0.09	3.49	1.65
cnt	BN	soil	2019	Harvest	7.57	1.59	13.78	4.74	0.84	0.43	0.03	0.10	3.04	1.88	0.00	0.00	4.24	1.27
zeo	BN	soil	2019	Harvest	7.40	1.29	15.81	7.71	0.51	0.52	0.04	0.08	2.28	1.31	0.07	0.14	3.21	0.75
cnt	BN	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid
zeo	BN	soil	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid	covid
cnt	BN	soil	2020	Post-Fert	0.31	0.10	30.92	41.88	0.25	0.31	0.00	0.00	22.60	15.96	0.00	0.00	4.79	3.21
zeo	BN	soil	2020	Post-Fert	0.33	0.12	5.34	2.93	0.22	0.10	0.00	0.00	29.50	5.43	0.00	0.00	4.88	2.02
cnt	BN	soil	2020	Harvest	3.62	0.38	40.85	22.76	0.82	0.33	0.00	0.00	54.80	61.89	0.09	0.15	11.06	8.10
zeo	BN	soil	2020	Harvest	3.29	0.35	21.75	10.96	5.69	5.86	0.00	0.00	199.86	139.03	0.09	0.16	19.61	10.17
cnt	BN	soil	2021	Pre-Fert	3.62	0.38	40.85	22.76	0.82	0.33	0.00	0.00	54.80	61.89	0.09	0.15	11.06	8.10
zeo	BN	soil	2021	Pre-Fert	3.29	0.35	21.75	10.96	5.69	5.86	0.00	0.00	199.86	139.03	0.09	0.16	19.61	10.17
cnt	BN	soil	2021	Post-Fert	4.78	1.15	32.73	13.16	6.44	4.72	0.00	0.00	413.59	260.51	0.00	0.00	21.05	19.29
zeo	BN	soil	2021	Post-Fert	4.21	0.84	46.33	16.91	12.41	5.33	0.00	0.00	975.43	456.03	0.00	0.00	49.05	42.54
cnt	BN	soil	2021	Harvest	3.10	0.85	18.87	0.91	0.22	0.04	0.00	0.00	31.83	24.17	0.00	0.00	9.72	7.21
zeo	BN	soil	2021	Harvest	2.84	0.74	33.42	24.93	16.24	27.88	0.00	0.00	44.69	31.58	0.32	0.31	7.42	1.55

Treatment	Area	Type	Year	Management_status	TC (%)	dev.std TC	TN (%)	dev.std TN	$\delta^{13}C$	dev.std $\delta^{13}C$	$\delta^{15}N$	dev.std $\delta^{15}N$
cnt	SL	leaf	2019	Pre-Fert	44.951	1.322	2.069	0.189	-26.822	1.852	2.164	0.757
zeo	SL	leaf	2019	Pre-Fert	46.353	0.384	2.020	0.230	-25.966	0.431	1.795	1.429
cnt	SL	leaf	2019	Post-Fert	49.979	3.420	1.968	0.218	-25.243	0.458	3.629	0.607
zeo	SL	leaf	2019	Post-Fert	46.048	1.535	1.778	0.036	-24.905	0.634	2.631	1.419
cnt	SL	leaf	2019	Harvest	46.427	2.959	1.665	0.290	-25.503	0.558	4.088	0.563
zeo	SL	leaf	2019	Harvest	51.354	3.069	1.833	0.145	-25.608	0.419	2.537	1.086
cnt	SL	leaf	2020	Pre-Fert	covid	covid	covid	covid	covi	covid	covid	covid
zeo	SL	leaf	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid
cnt	SL	leaf	2020	Post-Fert	46.673	4.178	1.672	0.228	-26.053	0.724	2.770	1.700
zeo	SL	leaf	2020	Post-Fert	46.350	2.138	1.739	0.198	-24.423	0.918	1.787	1.272
cnt	SL	leaf	2020	Harvest	47.580	0.574	1.407	0.327	-24.390	2.144	0.723	4.517
zeo	SL	leaf	2020	Harvest	47.320	2.696	1.927	0.194	-28.320	6.699	0.563	4.027
cnt	SL	leaf	2021	Pre-Fert	44.523	2.572	1.845	0.332	-25.170	0.667	1.857	1.287
zeo	SL	leaf	2021	Pre-Fert	44.027	3.639	2.002	0.161	-25.153	0.451	2.643	0.093
cnt	SL	leaf	2021	Post-Fert	37.440	8.073	1.487	0.239	-23.797	0.423	0.683	1.946
zeo	SL	leaf	2021	Post-Fert	35.660	4.966	1.453	0.125	-24.010	0.199	2.083	0.692
cnt	SL	leaf	2021	Harvest	57.773	14.392	2.215	0.562	-57.867	1.904	2.217	2.276
zeo	SL	leaf	2021	Harvest	47.517	3.843	1.872	0.170	-49.247	6.866	-0.313	3.204
cnt	BG	leaf	2019	Pre-Fert	46.909	2.163	1.960	0.421	-24.922	0.237	2.645	0.893
zeo	BG	leaf	2019	Pre-Fert	45.808	0.766	1.709	0.146	-24.763	0.571	3.452	1.264
cnt	BG	leaf	2019	Post-Fert	42.201	8.359	1.059	0.225	-24.498	0.243	1.506	0.384
zeo	BG	leaf	2019	Post-Fert	46.861	0.521	1.160	0.105	-24.323	0.134	2.452	0.898
cnt	BG	leaf	2019	Harvest	57.960	7.439	1.395	0.403	-26.670	3.380	1.060	0.028
zeo	BG	leaf	2019	Harvest	49.652	1.384	1.249	0.148	-33.567	15.090	1.185	0.807
cnt	BG	leaf	2020	Pre-Fert	44.525	1.294	0.982	0.095	-24.380	0.297	0.795	1.110
zeo	BG	leaf	2020	Pre-Fert	43.985	4.476	0.945	0.018	-24.635	0.304	1.065	1.817
cnt	BG	leaf	2020	Post-Fert	48.120	1.218	0.794	0.058	-25.527	2.324	-0.303	0.692
zeo	BG	leaf	2020	Post-Fert	42.583	16.094	-0.151	1.814	-24.943	1.262	0.650	0.996
cnt	BG	leaf	2020	Harvest	40.363	0.627	1.105	0.060	-27.173	2.032	1.360	2.512
zeo	BG	leaf	2020	Harvest	44.980	5.983	1.263	0.175	-32.517	13.963	0.523	1.900
cnt	BG	leaf	2021	Pre-Fert	43.330	5.012	1.280	0.203	-24.670	0.319	1.505	0.726

E

zeo	BG	leaf	2021	Pre-Fert	44.215	4.593	1.412	0.416	-24.723	0.393	0.565	1.013
cnt	BG	leaf	2021	Post-Fert	46.297	7.252	1.474	0.364	-24.023	0.352	-2.117	1.930
zeo	BG	leaf	2021	Post-Fert	50.340	4.946	1.600	0.303	-24.183	0.060	-0.623	0.822
cnt	BG	leaf	2021	Harvest	52.327	2.120	1.690	0.272	-35.057	14.939	-0.107	1.804
zeo	BG	leaf	2021	Harvest	61.913	7.955	2.029	0.498	-36.667	22.806	-0.350	0.984
cnt	BN	leaf	2019	Pre-Fert	44.740	2.019	2.169	0.256	-25.918	0.425	0.340	1.462
zeo	BN	leaf	2019	Pre-Fert	45.166	1.019	2.267	0.262	-26.086	0.391	0.009	1.822
cnt	BN	leaf	2019	Post-Fert	44.855	1.274	1.794	0.045	-25.401	0.406	-1.932	0.627
zeo	BN	leaf	2019	Post-Fert	46.023	0.544	1.926	0.088	-26.487	0.579	-1.769	0.644
cnt	BN	leaf	2019	Harvest	48.140	4.739	2.081	0.125	-36.776	19.224	0.177	1.318
zeo	BN	leaf	2019	Harvest	51.932	7.873	2.289	0.295	-28.944	3.285	1.494	1.386
cnt	BN	leaf	2020	Pre-Fert	covid	covid	covid	covid	covid	covid-	covid	covid
zeo	BN	leaf	2020	Pre-Fert	covid	covid	covid	covid	covid	covid	covid	covid
cnt	BN	leaf	2020	Post-Fert	53.300	4.775	1.739	0.201	-29.220	7.838	-1.667	1.358
zeo	BN	leaf	2020	Post-Fert	47.937	2.336	1.721	0.150	-25.370	0.661	-1.627	0.248
cnt	BN	leaf	2020	Harvest	47.993	0.842	1.817	0.300	-24.560	0.439	-1.553	0.903
zeo	BN	leaf	2020	Harvest	49.257	2.115	2.025	0.269	-25.657	1.831	-0.790	1.502
cnt	BN	leaf	2021	Pre-Fert	47.993	0.842	1.817	0.300	-24.560	0.439	-1.553	0.903
zeo	BN	leaf	2021	Pre-Fert	49.257	2.115	2.025	0.269	-25.657	1.831	-0.790	1.502
cnt	BN	leaf	2021	Post-Fert	50.277	6.019	2.122	0.459	-23.813	0.520	-0.317	0.345
zeo	BN	leaf	2021	Post-Fert	48.227	13.246	1.910	0.493	-24.320	0.707	-2.377	1.756
cnt	BN	leaf	2021	Harvest	48.630	4.241	1.927	0.043	-33.620	14.441	-1.130	2.082
zeo	BN	leaf	2021	Harvest	50.920	5.931	1.996	0.383	-45.230	19.645	-0.990	1.428