



# Innovative Tools for the Nitrogen Fertilization Traceability of Organic Farming Products

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**Abstract:** In the last decades, consumers have become increasingly interested in organic products, and they strongly demand reliability in the traceability of the organic products they buy and eat. Several research methods have been developed in the last decades to study inexperienced and reliable systems and to assess the authenticity of products obtained using organic cultivation practices. The monitoring of some chemical compounds, originating from primary and/or secondary metabolism, in horticultural organic and conventional products has shown the diversity generated using the two production approaches. The difference in fertilization practices has been also shown to have an effect on the isotopic distribution of some elements, with particular reference to nitrogen. An integrated system is proposed to evaluate the validity of organic goods using collected isotopic data and other chemical and biological parameters. This approach is intended to be coupled to the application of chemometric multivariate analysis on quality and nutraceutical parameters combined with isotopic data. Indeed, this will give the opportunity to discriminate organic from conventional products based on different isotopic signatures, due to the different nitrogenous sources, combined with the qualitative profile of the crops, which are significantly affected by the different agronomic treatments. The main perspectives of the presented integrated approach, based on the combined use of chemometric and analytical tools, are linked to the feasibility of applying a reliable system for traceability. This will authenticate productions obtained using organic fertilizers (organic agriculture) with respect to those obtained with the use of synthetic fertilizers (conventional agriculture), protect and valorize virtuous farmers and support political stakeholders and decision-makers to counteract food fraud.

**Keywords:** organic horticultural practices and production; chemical metabolites; isotopic ratios; multivariate analysis; chemometrics



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## 1. Background

In recent years, a growing interest in organic products has been recorded as they are considered more sustainable, safer, and healthier than conventional ones by consumers. As a matter of fact, the traceability of organic products and the definition of their authenticity have been subjects of relevant interest for many years. Several studies have been performed to investigate the authenticity of products obtained using organic cultivation methods [1,2]. They focused on the identification of new quality “markers” that allow

differentiating, from field to fork, an organic product from a conventional one. In particular, the monitoring of some chemical compounds, originating from primary and/or secondary metabolism, in horticultural organic and conventional products has shown the diversity generated using the two production approaches [3,4]. Indeed, based on biochemical and transcriptional approaches, crucial evidence has been provided about the physiological changes that can occur in organic and conventional cultivated crops [5]. Moreover, it is widely recognized that the diversity in fertilization practices for the two cultivation systems influences the isotopic distribution of some elements present in fruit and vegetable crops, with particular reference to nitrogen (N) [6,7]. Starting from these backgrounds and the knowledge shared so far within the scientific community, the main aim of the present communication is to propose an innovative approach for the N fertilization traceability of organic farming products.

## 2. The Up-to-Date Literature

Based on the Sustainable Development Goals introduced by the United Nations [8] and the targets of the European Green Deal [9], it is known that one of the main worldwide shared goals is to increase the percentage of organically managed farms by 2030. The focused target is to have 25% of the whole European cultivated area devoted to organic agriculture. In this context, it has been widely discussed how organic agriculture is more environmentally sustainable with respect to conventional farming for different reasons, including lower nutrient losses, reduced fossil energy consumption, and the ban on synthetic pesticides and fertilizers [10]. One of the most important aspects of organic agriculture is to manage N fertilization since synthetic fertilizers are not allowed. In fact, N is a crucial element for plant growth, and it has been estimated that about half of the amount of N provided to the soil is lost [11]; therefore, there is a need to synchronize the amount of N demand and the N supply when different fertilization strategies are used, specifically when organic and conventional practices are used [12]. Furthermore, consumers are increasingly interested in organic food, and they strongly demand reliability in the traceability of the organic products they usually buy and eat. The actual European regulation (EU Reg. 2018/848) establishes that official controls are to be performed in accordance with Regulation (EU) 2017/625 (art. 14) [13]. Specifically, this regulation establishes that official control methods and techniques shall include the examination of documents and traceability records that may be relevant to an assessment for compliance with the rules of organic management. In fact, on the majority of organic farms, the sole traceability mechanism in place is based on documentary evidence that agricultural management complies with European regulations. Furthermore, multiresidue analyses are carried out only upon the specific request from certification bodies or distribution buyers, and relative samplings are randomly performed [14,15]. Indeed, this might not be enough to certify the authenticity of organic products as farmers could fraudulently use fertigation to supply mineral fertilizers to the soil or apply pesticides, observing large shortage intervals, and all these behaviors would not be easily detected [16,17]. So, there is a need for an objective traceability of organic management based on scientifically collected data [18].

The possibility of differentiating two samples on the basis of isotopic analysis is linked to the isotopic fractionation that occurs in raw materials. Isotopic ratios of natural elements such as nitrogen, carbon, oxygen, hydrogen, and sulfur have been conveniently used for food authenticity purposes, as their fractionation occurs in response to very different biogeochemical phenomena [19]. It has been demonstrated that isotopic analysis can be reliably used to check for food authenticity [20–22]. Specifically for organic products, a deviation from the natural isotopic distribution of N happens during the production and maturation of organic fertilizers. Indeed, enrichment in the lighter isotope ( $^{14}\text{N}$ ) occurs due to its preferential volatilization. As a consequence, the N isotopic ratio [ $\delta(^{15}\text{N}) = ^{15}\text{N}/^{14}\text{N}$ ]

$$\delta^i \left( E_{\text{sample/standard}} \right) = \frac{R(i_E/j_E)_{\text{sample}}}{R(i_E/j_E)_{\text{standard}}} - 1$$

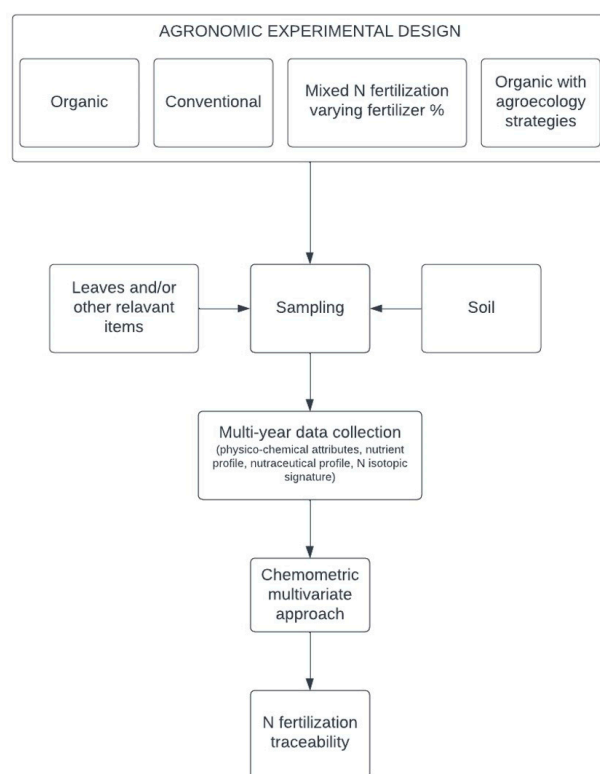
is increased in organic fertilizers and amendments of both vegetable and animal origin [6,7]. In the aforementioned equation,  $\delta(^{15}\text{N})$  is expressed with respect to the international standard air (atmospheric  $\text{N}_2$ ), and  $i_E/j_E$  is the isotope ratio between the heavier and lighter N isotopes. Air has more or less the same N isotopic composition throughout the world ( $\delta(^{15}\text{N}) = 0\text{‰}$  by definition and internationally recognized), and industrially produced mineral fertilizers typically have  $\delta(^{15}\text{N})$  close to 0‰ because their N directly derives from atmospheric nitrogen, and there is basically no isotope fractionation during their industrial synthesis [6,7]. Conversely, organic fertilizers allowed in organic farming have positive values of  $\delta(^{15}\text{N})$  (ranging from +1‰ to +37‰); therefore, they are easily distinguishable from synthetic fertilizers. It has been demonstrated that these differences are reflected in the final vegetable product, thereby confirming the great potential for N isotopic analysis to check the authenticity of organic products [23–33]. At the same time, it must be stressed that it has been widely argued that  $\delta(^{15}\text{N})$  may not be discriminating for all crops or in every management condition, e.g., when organic fertilizers are contemporarily used in conventional practice, and vice versa, or for crops which are characterized by low N requirements [33]. In addition, it is also worth noting that few studies have been reported on the effect of using agricultural service crops (such as N-fixing plants (legumes) or not N-fixing plants) for green manure in organic management on the isotopic signature of the final product [34]. As a matter of fact, N-fixing plants fix nitrogen from the air (having  $\delta(^{15}\text{N})$  values close to 0‰) in root nodules, thus the N isotopic signature of the final products could reflect this root uptake, putatively leading to misclassification of authentic organic products.

Several researchers have reported that organically grown fruit is richer in antioxidants when compared with conventional counterparts [3,35–37]. For almost two decades, it has been postulated that N deficiency in organic products may induce the production of carbon-rich phenylpropanoids, thus explaining the higher healthy value of organic products. Recent research [4] has reported results that conflict with this theory, showing that long-term organic fields are associated with no relevant or significant difference in N and carbon (C) content or in the C:N ratio between organic and conventional products. The same study reported that pest attacks were preferentially set on conventional plants with respect to organic ones, but no evidence suggesting plant nutrient unbalance was directly linked with this result. Consistently, the study demonstrated that organic soil management boosted salicylic acid build-up, which resulted in discouraging plant–insect interactions. Indeed, salicylic acid accumulation was not associated with N deficiency in the organic plants, but it depended on adaptations in soil microbial communities associated with long-term organic management. Thus, based on this recent experimental evidence reporting how soil microbiota influences plant resistance to pests, it may also be postulated that polyphenolics and other antioxidants may accumulate in organic plants due to functional shifts in soil microflora. Furthermore, based on a biochemical and molecular approach, it was recently demonstrated [5] that different fertilization practices (organic or conventional) induced differential accumulation of free amino acids, with higher concentrations of proline and contemporarily lower concentrations of glutamate in fruits from organically managed orange trees. In addition, the upregulation of glutamate dehydrogenase gene expression was recorded, thus identifying a possible adaptive response in common orange plants to organic fertilizers. It has been suggested that this adaptive response may putatively be based on a metabolism switch aimed at ensuring turnover of the tricarboxylic acid cycle to continuously supply carbon skeletons for biosynthetic demands, as a regulatory phenomenon. Other studies recently reported the possibility of discriminating between organic and conventional crops based on the differential metabolome associated with the two productions [38,39].

### 3. Methodology

In order to achieve the goal of implementing a reliable system for tracing the authenticity of organic products, different treatments should be used and compared. One

possibility is to incorporate conventional, organic, and mixed N fertilization options in the experimental design, comparing them at varying fertilizer percentages. It is also relevant to include the use of agronomic techniques for the management of soil fertility, such as the use of agroecology strategies [40]. The variations in  $\delta(^{15}\text{N})$  induced using the combination of different fertilizations together with the use of green manure or cover crops exhibiting an agroecological function in the organic production system should be quantified. This would allow for evaluating how soil management based on the agroecological approach affects the  $\delta(^{15}\text{N})$  in the final product, in comparison with organic management based on the full substitution and/or conventional agriculture approaches. At the same time, the variations in physicochemical and nutraceutical attributes induced using the different treatments should be evaluated. Once collected, the obtained data should be treated with a chemometric multivariate approach able to display the data in a way that chemical interpretation of the system is allowed. This may involve transforming the original variable into new variables that are functions of the original data and verifying that a sample can be assigned to previously defined groups formed using samples of known origin ('organic' or 'not organic') on the basis of their compositional similarity with groups. This system will be able to discriminate, at a high confidence level, between organic and conventional products. Finally, the use of the implemented database should be validated on a multiyear basis (Figure 1).



**Figure 1.** Methodology flow chart.

#### 4. Perspectives

The main aim of the present communication is to provide insights and general suggestions for the application of a feasible methodology for the implementation of an investigation system based on a combined chemical and chemometric approach. In particular, the main perspectives for the application of this kind of approach are linked to the possibility to: (i) allow, with the acquisition of relevant chemical and biochemical attributes and isotopic data, one to discern between productions obtained with mineral fertilizers, typically applied in conventional agriculture and not allowed in organic agriculture, and productions obtained with organic agriculture practice; (ii) improve the general under-

standing of the factors influencing the quality of organic products and their differentiation from conventional ones; (iii) allow the traceability of organic products from field to fork which, in turn, would contribute to protect and reward virtuous farmers; (iv) valorize the environmental externalities linked to the application of organic farming practices; and (v) support policymakers in identifying methodologies for contrasting food fraud.

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