**Introduction**

The growing demand for organic produce has increased the need for robust analytical methods to verify labeling and trace product origin. Stable isotope ratio analysis has emerged as a powerful approach in food authentication, since the isotopic composition of plants reflects both nutrient inputs and environmental conditions (Kelly et al., 2005). Among these, carbon (δ¹³C) and nitrogen (δ¹⁵N) isotopes are particularly relevant to agricultural systems and are increasingly applied in food authenticity studies (Bontempo et al., 2020).

Nitrogen isotopes provide insight into fertilizer sources. Synthetic nitrogen fertilizers produced via the Haber–Bosch process generally have δ¹⁵N values near 0‰, while organic amendments such as manures and composts are enriched in ¹⁵N (due to microbial processing ) and volatilization (Bateman & Kelly, 2007). Consequently, crops grown with organic fertilizers often show higher δ¹⁵N values than those cultivated with synthetic inputs. Tomato studies confirm this pattern: (Nakano et al., 2003) observed δ¹⁵N of +7.1‰ under organic fertigation versus +0.3‰ with inorganic fertilizer, and (Trandel et al., 2018) reported consistently higher δ¹⁵N in greenhouse tomatoes amended with organic matter. However, our findings align with this pattern, showing a highly significant enrichment of +5–6‰ in organic tomato products relative to conventional ones, with almost no overlap between groups. This demonstrates δ¹⁵N as a robust discriminator of production method. In contrast, δ¹³C is less effective for distinguishing farming systems, since it mainly reflects plant water-use efficiency and environmental stress rather than fertilizer type (Nakano et al., 2003; Bontempo et al., 2020). We also found similar results in our study, δ¹³C differences by farming type were marginal and only regionally consistent (e.g., Bavarian samples), highlighting environmental drivers as the main source of variability.

Beyond production methods, stable isotopes also carry regional signals. Climate, soil type, and hydrology can alter baseline isotope values, independently of farming system (Amundson et al., 2003). Multi-isotope approaches have been successful in distinguishing regional origins of foods: (Bontempo et al., 2011) achieved high accuracy in classifying Italian tomato products using δ¹³C, δ¹⁵N, δ³⁴S, and mineral profiles, while Kelly et al. (2005) highlighted the value of isotopes in geographic traceability across diverse commodities. Yet, in our dataset, bulk δ¹³C and δ¹⁵N achieved only ~65% accuracy for regional classification, underscoring their limited power for origin tracing when used alone. δ¹³C clearly separated more arid vs. humid regions, while δ¹⁵N varied with both fertilizer type and soil baselines, producing partial confounding of farming and regional effects, perhaps due to imbalanced sampling. Moreover, as Bontempo et al. (2020) showed for tomato passata, bulk δ¹³C and δ¹⁵N alone provide strong discrimination of farming method but limited power for clear regional separation, underscoring the importance of compound-specific or multi-isotope approaches.

We therefore address two overarching questions: (1) Can δ^15N and δ^13C reliably differentiate tomato products by farming type across different regions? (2) To what extent is the isotopic signature driven by production method versus geographic/environmental factors? By sampling tomato fruits and derived products from multiple regions and cultivation systems, this study evaluates isotope differences under real-world conditions and discusses the potential and limits of δ^15N and δ^13C as markers of organic provenance.

Materials and Methods

**Sample collection and preparation**

Tomato products (A–J) representing both organic and conventional farming systems were purchased from major European retailers (Aldi, Kaufland, Ebl Naturkost, and EDEKA). Samples originated from three main production regions: Bavaria (Germany), Almería (Spain), and Souss-Massa (Morocco). Each product was subsampled in quadruplicate (except Bn=12, Dn=3, and Jn=9), n = 52 total. All samples were freeze-dried, homogenized with a mortar and pestle, and stored airtight. Approximately 1000 µg ± 100 µg of homogenized powder was weighed into tin capsules (IVA, Germany) for analysis.

**Isotopic and elemental analysis**

Stable isotope ratios of carbon (δ¹³C) and nitrogen (δ¹⁵N), together with elemental composition (%C, %N, and C:N ratio), were analyzed using a Flash 2000 Elemental Analyzer (Thermo Scientific) coupled via a Conflo II interface to a Delta V Advantage IRMS (Thermo Scientific). Isotope values are expressed in δ notation (‰) relative to Vienna Pee Dee Belemnite (VPDB) for carbon and atmospheric N₂ (AIR) for nitrogen. Calibration was performed against international reference standards (IAEA-N1, IAEA-N2, USGS-41, IAEA-CH7) and a laboratory standard (“Peptone II”), with elemental quantification calibrated using atropine and cyclohexanone-2,4-dinitrophenylhydrazone.

**Statistical analysis**

All analyses were performed in R (v4.4.2). Farming system comparisons were evaluated with Welch’s t-tests (reporting t, df, and p). Regional effects were tested using one-way ANOVA, while combined two-way ANOVA models assessed the influence of Region, Farming Type, and their interaction. For ANOVAs, F-statistics, p-values, and η² effect sizes were extracted. Multivariate structure was explored using principal component analysis (PCA) on z-standardized isotopic and elemental variables (δ¹³C, δ¹⁵N, %C, %N, C:N), and classification performance was assessed using linear discriminant analysis (LDA). PCA variance explained by the first components and LDA classification accuracies for farming type and region were reported in the Results. Statistical significance was set at α = 0.05.