

Development of a HS/GC-IMS Screening Method for Honey Geographical Origin Classification

Giancarlo Quaglia¹, Cesare Rossini² & Matteo Bordiga³



¹ Department of Drug Science and Technology, University of Torino, Torino, Italy

² Lab Service Analytica s.r.l, Via Emilia 51/c, 40011 Anzola Emilia (Bologna), Italy

³ Department of Pharmaceutical Sciences, University of Piemonte Orientale, Novara, Italy

matteo.bordiga@uniupo.it



CONTEXT

In recent years, the geographical provenance of honey has become a salient issue for consumers and regulatory authorities, as it exerts a significant influence on the perception of the product's quality, authenticity and economic value. The recent revision of the Honey Directive, as part of the European Union's so-called 'Breakfast Directives', further reinforces this principle. The revision introduces stricter requirements regarding origin labelling, mandating the indication of the countries of harvest and their respective percentages in blends, as well as providing for future harmonized traceability systems and analytical methods to support authenticity verification. These changes underscore the necessity for expeditious, dependable, and economical analytical instruments to ascertain the congruence between the declared and actual origins of honey. At present, the determination of honey's geographical origin is based primarily on melissopalinalogical analysis and advanced analytical techniques, such as nuclear magnetic resonance (NMR) spectroscopy, metabolomics using high-resolution mass spectrometry, and multivariate chemometric approaches. Despite the fact that these methodologies deliver high analytical performance, they necessitate costly equipment, highly qualified staff, and analysis times that are not always compatible with large-scale routine testing [1, 2].

In this context, the use of ion mobility spectrometry (IMS), utilizing FlavourSpec® instrumentation, represents a promising alternative for the development of high-throughput, low-cost screening methods. IMS technology facilitates the expeditious acquisition of a volatile fingerprint of honey, which exhibits a high degree of correlation with its aromatic composition. This composition is, in turn, influenced by the botanical and geographical characteristics of the production area.

The integration of IMS data with multivariate analysis tools and classification algorithms facilitates the effective differentiation of samples from disparate geographical areas, thereby enabling an initial assessment of the product's provenance within a brief timeframe and at a substantially reduced analytical cost when compared to confirmatory methods.

HS-GC-IMS: Samples of honey from four different geographical origins (Brazil, China, Hungary and Chile) were used. Headspace-gas chromatography-ion mobility spectrometry (HS/GC-IMS) (FlavourSpec®, G.A.S., Dortmund, Germany) was used to assess the volatile composition with an untargeted fingerprinting approach. A 20 mL glass vial was filled with 1.0 g of the sample. Then samples were treated for 5 minutes at 40 °C at 500 rpm. Then, in splitless mode, a 300 µL headspace sample was automatically delivered through a 50 °C heated syringe. Using an MXT-5 column (15 m × 0.53 mm i.d., 1 µm film thickness; Restek Corporation, Bellefonte, PA, USA), the volatile chemicals were separated at 40 °C. As the carrier gas, 99.999 percent pure nitrogen was employed, and the flow rate program was configured as follows: 2 mL/min for 2 minutes, followed by a 5-minute rise to 150 mL/min and a 3-minute hold. A 3H ionization source ionized the eluted analytes before driving them to a drift tube, which was run at a constant temperature of 45 °C and voltage of 5 kV.



The aim: To meet stricter European regulatory requirements for origin transparency, this study evaluated HS/GC-IMS (FlavourSpec®) combined with chemometric analysis as a high-throughput screening method for honey authentication. The approach successfully discriminated international honey samples across different geographical origins (Brazil, China, Chile, and Hungary) and botanical varieties by mapping their unique volatile organic fingerprints. Ultimately, this method serves as a rapid, cost-effective preliminary filter for official and in-house quality controls, effectively isolating suspect samples to protect consumer trust and combat commercial fraud.

RESULTS AND DISCUSSION

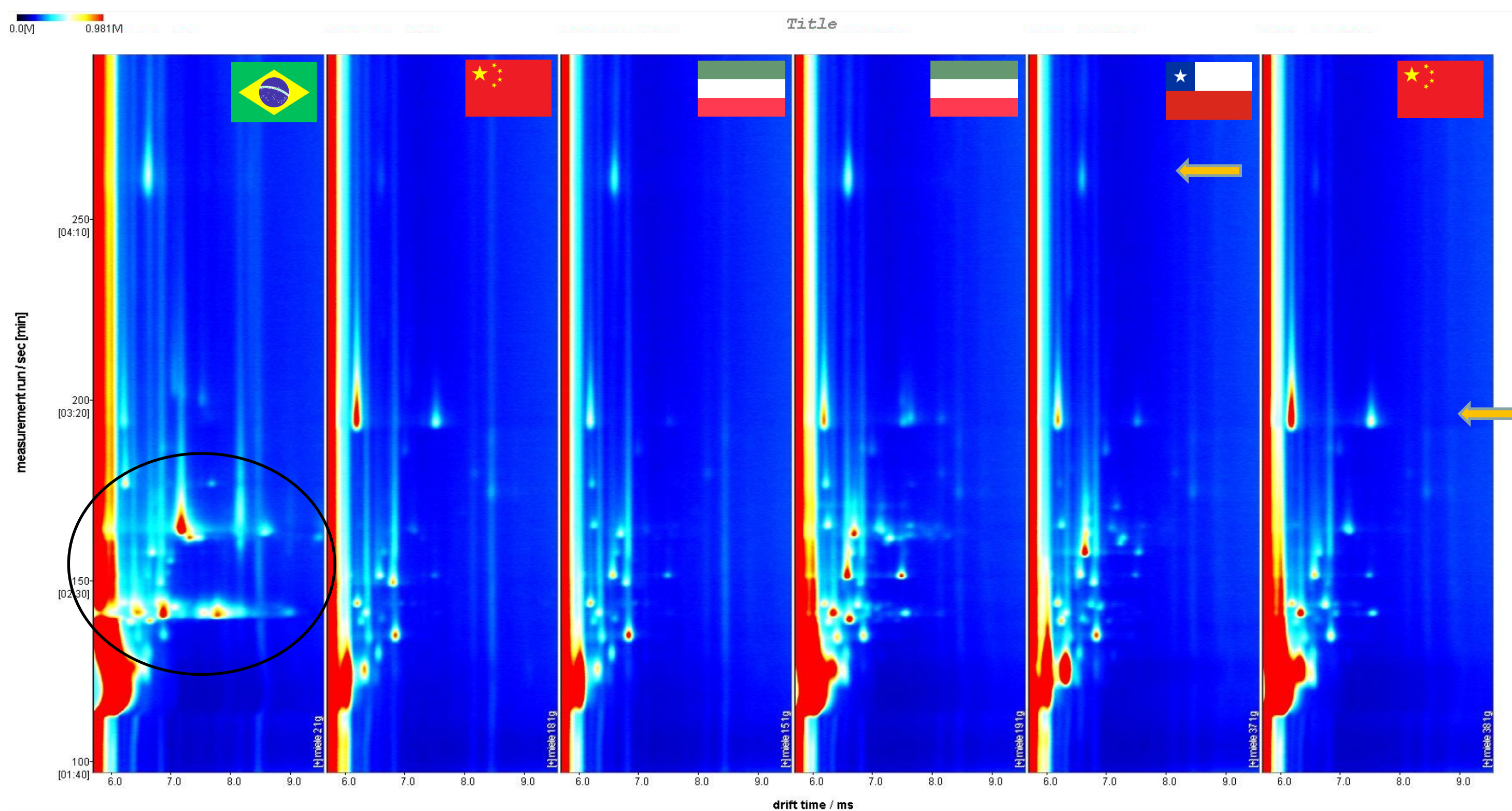


Figure 1. Two-dimensional chromatographic profiles of different types of honey.

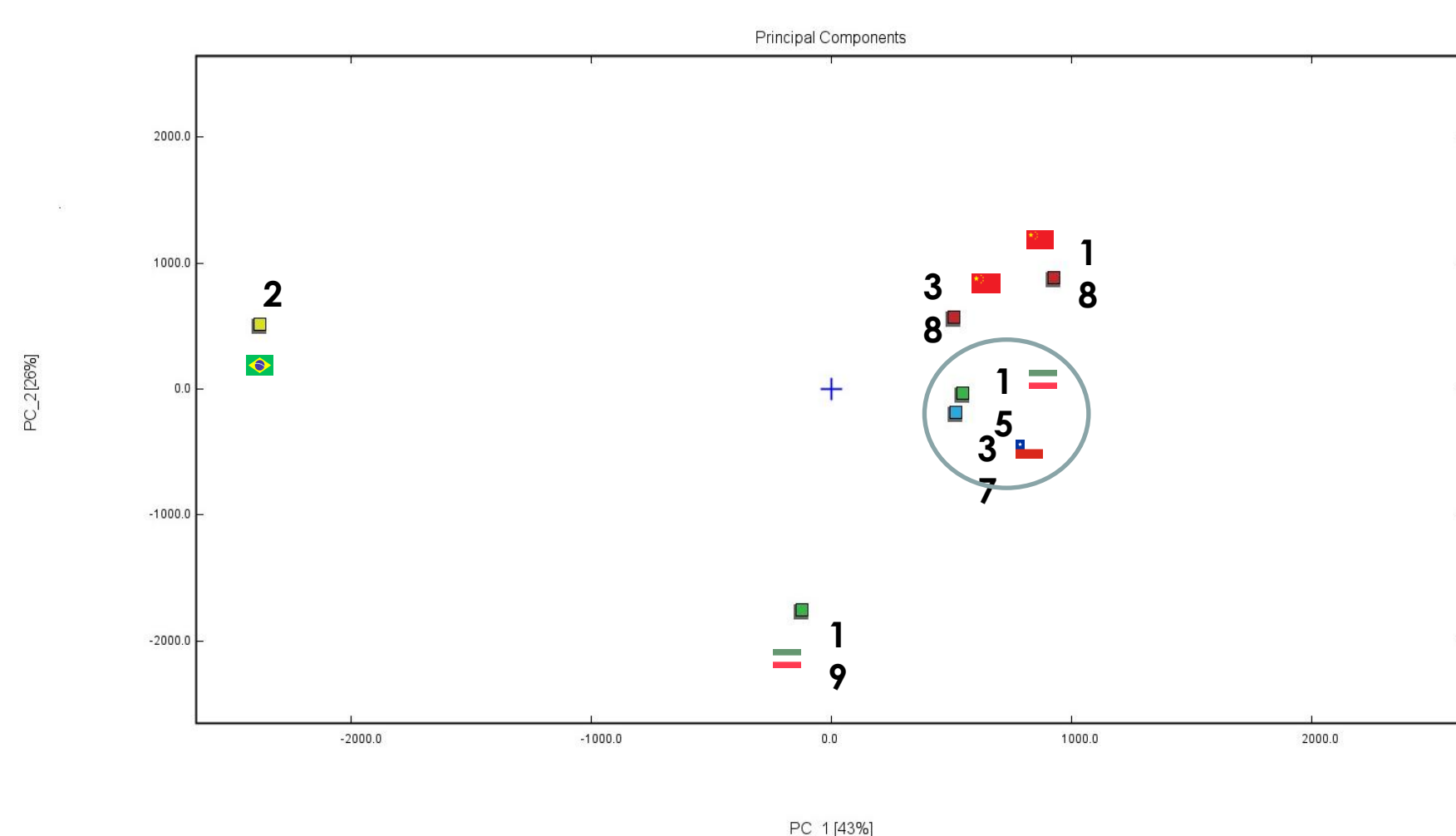


Figure 2. Principal Component Analysis (PCA) score plot of different honey samples based on volatile profiles.

Characterization of monofloral vs. polyfloral honey profiles by GC-IMS. The 2D topographical plots (retention time vs. drift time) demonstrate significant variations in volatile organic compounds (VOCs) highlighted by the white ellipses, proving the effectiveness of untargeted GC-IMS analysis for floral origin differentiation (Fig 3).

This study reports the volatile organic compound (VOC) analysis of six international honey samples analyzed using HS/GC-IMS. By combining gas chromatographic separation (retention time) with ion mobility drift time separation, GC-IMS generates two-dimensional "chemical fingerprints" of volatile profiles (Fig. 1). This technique enables non-destructive, highly sensitive authentication of geographical origin and differentiation between botanical varieties (Monofloral vs. Polyfloral) without extensive sample preparation. This study aimed to establish an accurate and robust screening method to classify honey's geographical origin for official and internal quality controls. Functioning as a high-throughput preliminary filter, it reduces costs by sending only suspect samples to complex confirmatory testing - ultimately supporting Food Information Directive standards, protecting consumers, and combating commercial honey fraud. Principal Component Analysis (PCA) explains 69% of total variance (PC1 = 43%, PC2 = 26%) and successfully segregates the samples into four distinct clusters (Fig 2).

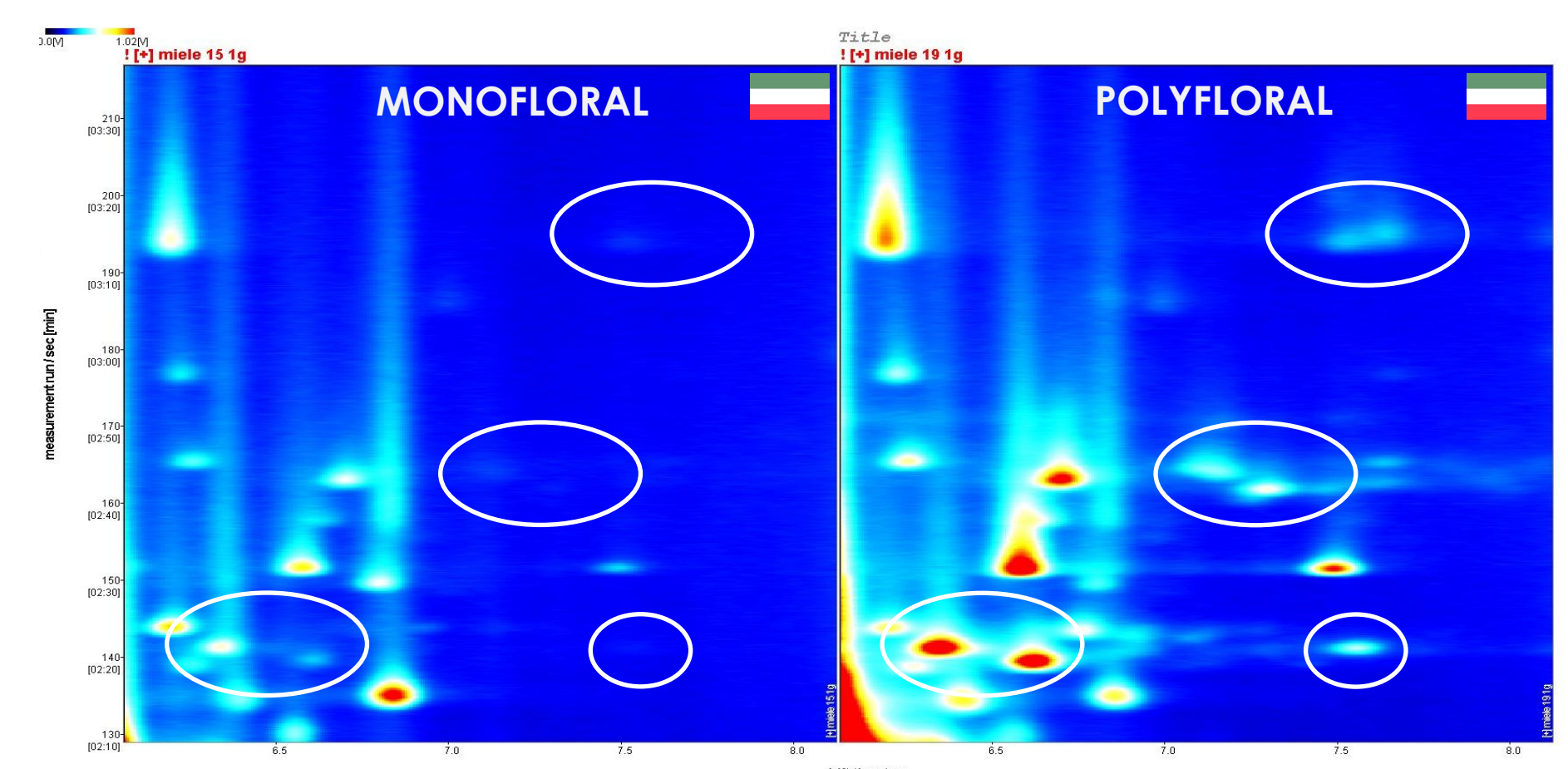


Figure 3. GC-IMS fingerprinting of monofloral and polyfloral honey.

CONCLUSIONS

This HS/GC-IMS screening approach seamlessly aligns with the evolving European regulatory framework. It effectively separates honeys by both country of origin and floral type, providing a reliable methodology for quality control, traceability, and fraud detection in commercial honey markets. By providing a fast, harmonized analytical tool, it strengthens origin verification and safeguards honey authenticity throughout the entire supply chain.

References

[1] Joscha Christmann, Sascha Rohn, Philipp Weller. Data in Brief 54 (2024) 110532.

[2] Hannah Schanzmann, Alexander L. R. M. Augustini Daniel Sanders, Moritz Dahlheimer, Modestus Wigger, Philipp-Marius Zech and Stefanie Silemann. Molecules 2022, 27, 7554.

