

Introduction

Almost 1000 compounds have been identified in roast coffee extracts, with chemical composition varying due to a range of factors, such as coffee bean origin and degree of roasting. The overall flavour and aroma results from a complex combination of chemical classes, including terpenes, oxygenates (aldehydes, esters and ketones) and thiophenes, as well as a range of nitrogen-containing compounds (pyrazines, pyridines and thiazoles).

Comprehensive two-dimensional gas chromatography coupled with time-of-flight mass spectrometry (GC×GC–TOF MS) is ideal for the analysis of such complex samples. The enhanced separation capacity of this technique allows characterisation of the entire sample in a single run.

In this study, key flavour compounds that would have been subject to extensive co-elution in a conventional GC–MS system were quickly and confidently identified using automated search tools and compared across multiple samples.

Experimental

Analysis of tea aroma

Sample preparation: Dried tea leaves (~100 mg) were placed inside an empty glass TD tube for direct desorption, enabling automation, while eliminating time-consuming sample preparation and the use of solvents.

TD method (TD-100™): Desorption: 80°C for 10 min with a 50 mL/min trap flow. Analytes trapped using an 'Odour' trap. Overall split 76:1.

GC×GC: Primary column: BPX5 (30m × 0.25 mm × 0.25 μm); Secondary column: BPX50 (2 m × 0.1 mm × 0.1 μm). Modulator: Zoex ZX2 loop modulator; Delay loop: 1 m × 0.1 mm × 0.1 μm (as for second column). Modulation period: 6 s. Main oven: 40°C (3 min), 4°C/min ramp to 260°C (10 min). No secondary oven offset.

TOF MS: BenchTOF-HD™ (Markes International); Transfer line: 250°C; Ion source: 250°C; Mass range: m/z 35–500; Data rate: 50 Hz.



Figure 1: TD-100 tray with glass tubes containing tea leaves.

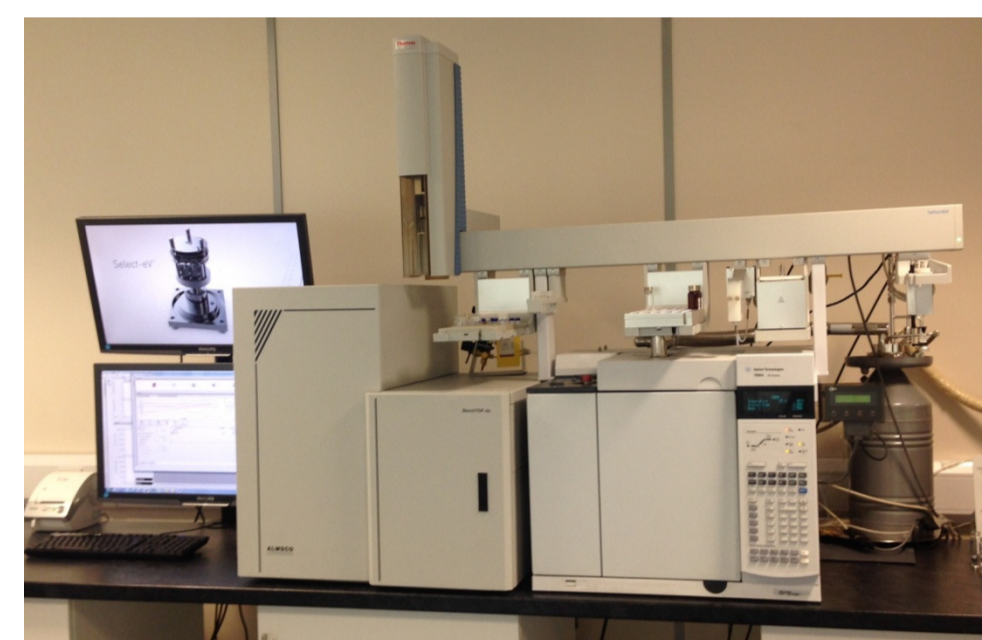


Figure 2: BenchTOF-HD coupled to GC×GC.

Analysis of coffee extracts

Sample preparation: Three roasted green coffees were analysed. Brewed coffee was neutralised with soda and extracted with dichloromethane.

GC×GC: Primary column: DB-50 (30m × 0.25 mm × 0.25 μm); Secondary column: Stabilwax® (0.6 m × 0.1 mm × 0.1 μm). Modulator: Zoex ZX1 loop modulator; Delay loop: 1 m × 0.1 mm × 0.1 μm (as for second column); Modulation period: 6 s. Main oven: 40°C (1 min), 3°C/min to 250°C (10 min). No secondary oven offset. Split 5:1.

TOF MS: Transfer line: 250°C; Ion source: 250°C; Mass range: m/z 35–400; Data rate: 50 Hz.

Results – Aroma profiling of tea

A selection of tea samples, including oolong and herbal varieties, were analysed by direct desorption TD–GC×GC–TOF MS. The resulting GC×GC colour plots are shown in Figure 3.

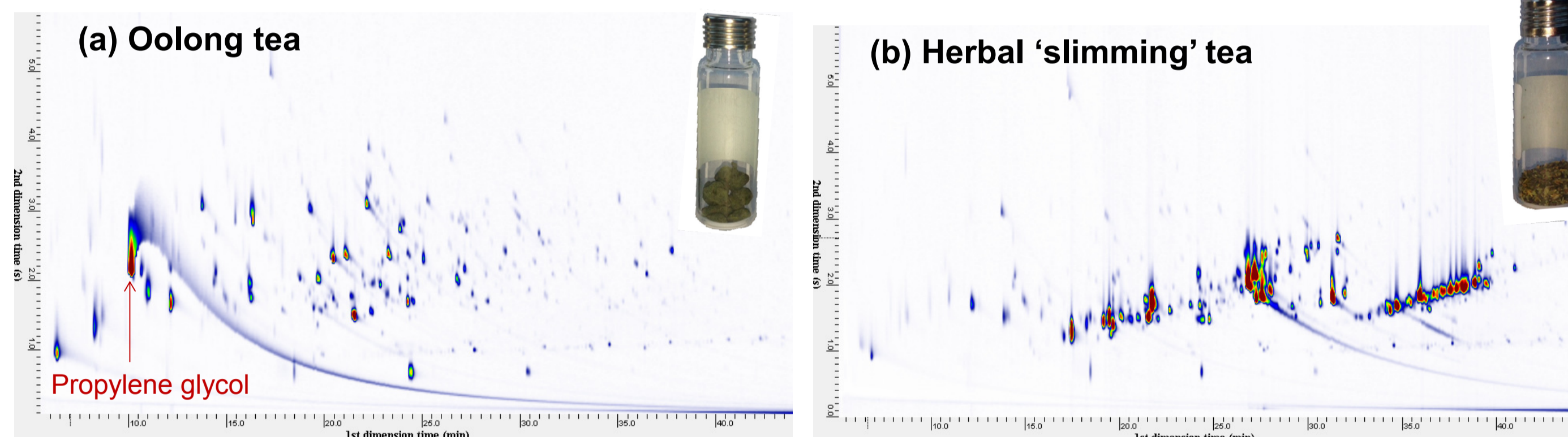


Figure 3: GC×GC–TOF MS colour plots for direct desorption of (a) oolong and (b) herbal 'slimming' tea.

The oolong tea was found to contain high levels of propylene glycol, which is used as a binding agent in the formation of the oolong pellets. The herbal tea, on the other hand, contained mainly mono- and sesquiterpenes and terpenoids (Figure 4).

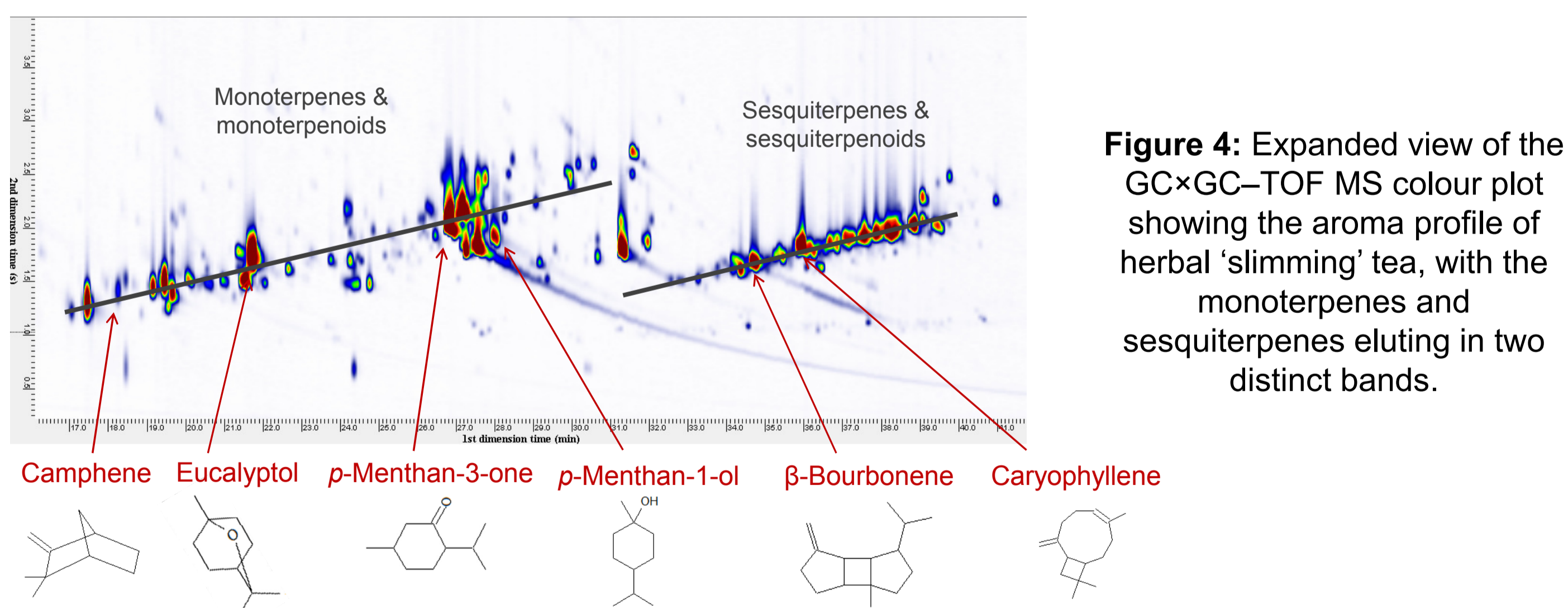


Figure 4: Expanded view of the GC×GC–TOF MS colour plot showing the aroma profile of herbal 'slimming' tea, with the monoterpenes and sesquiterpenes eluting in two distinct bands.

BenchTOF mass spectrometers use an ion source floating at 3 kV to minimise differences in ion impact velocities at the detector. This ensured that classical EI spectra were obtained for direct comparison to the NIST 14 database, as shown in the examples in Figure 5.

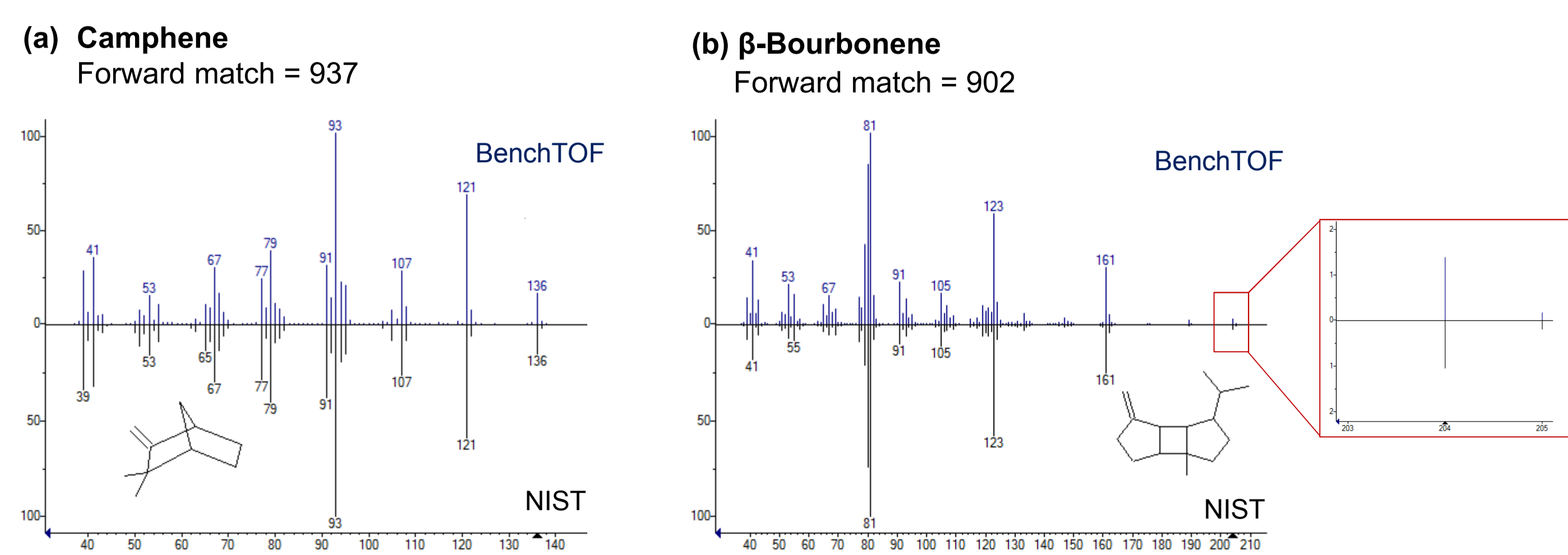


Figure 5: Spectral comparisons for (a) camphene and (b) β-bourbonene against the NIST 14 library.

Results – Characterisation of coffee

The coffee samples were analysed using a semi-polar to polar column set for optimal separation of the various polar constituents, including numerous classes of heterocyclics. The resulting GC×GC–TOF MS colour plots for the three coffee extracts are displayed in Figure 6.

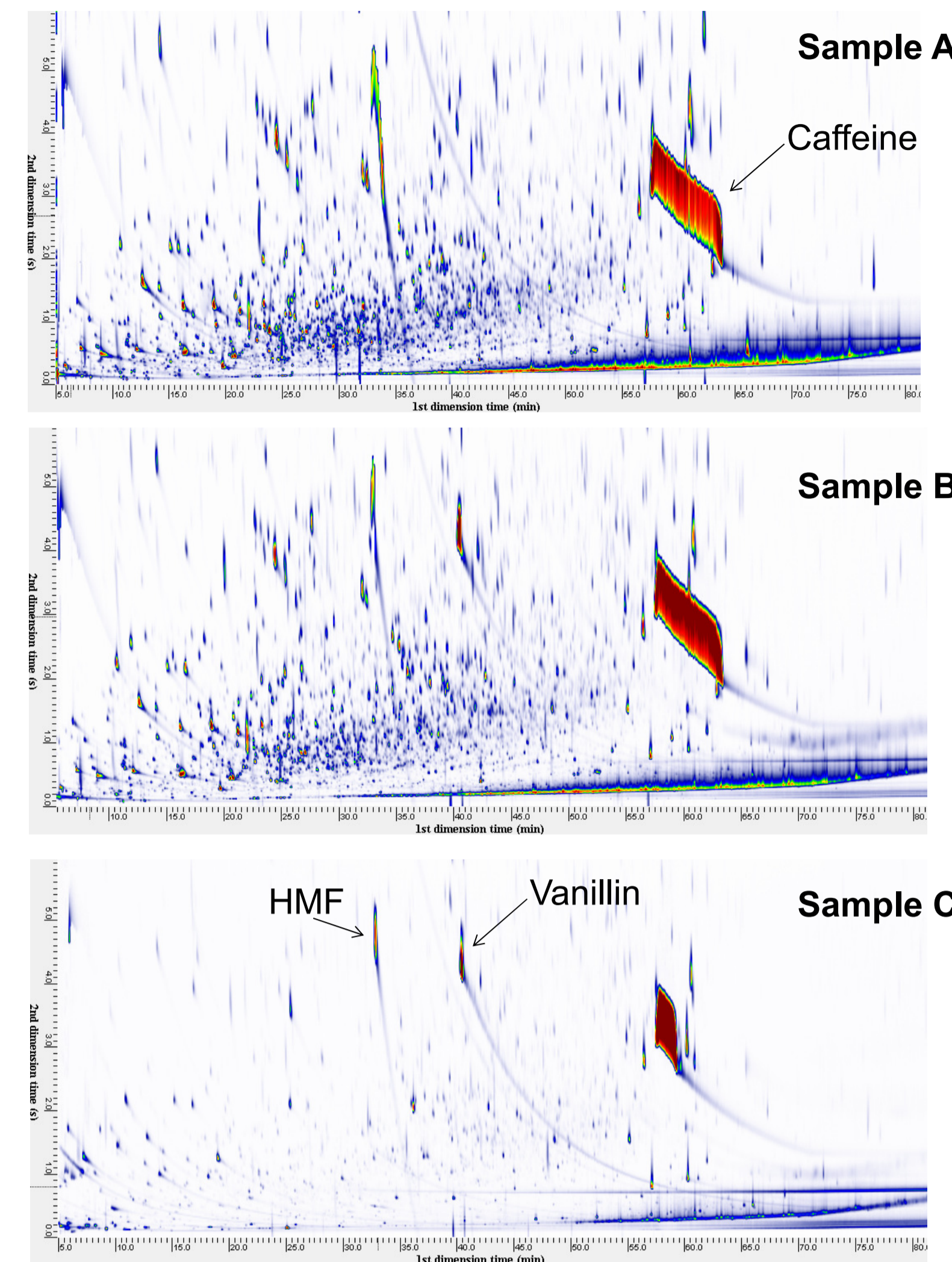


Figure 6: GC×GC–TOF MS colour plots for three coffee extracts.

As expected, the caffeine peak was the most prevalent in each of the three samples, with vanillin and 5-hydroxymethylfurfural (HMF) also in high abundance.

It is clear that samples A and B are similar, while sample C has a considerably less complex composition.

The extreme complexity of samples A and B would have resulted in numerous coelutions in conventional GC–MS analyses, but GC×GC–TOF MS ensures that as much information as possible is collected, enabling more confidence in product quality.

A simple scripting function was applied to identify the key compounds which contribute highly to the taste and aroma of coffee. For example, Figure 7 shows the pyrazines, pyridines and thiazoles that were found in Sample A based on their characteristic mass spectral fragmentation patterns. This enabled fast characterisation of the samples and easy incorporation of the scripts into a template to apply to subsequent samples.

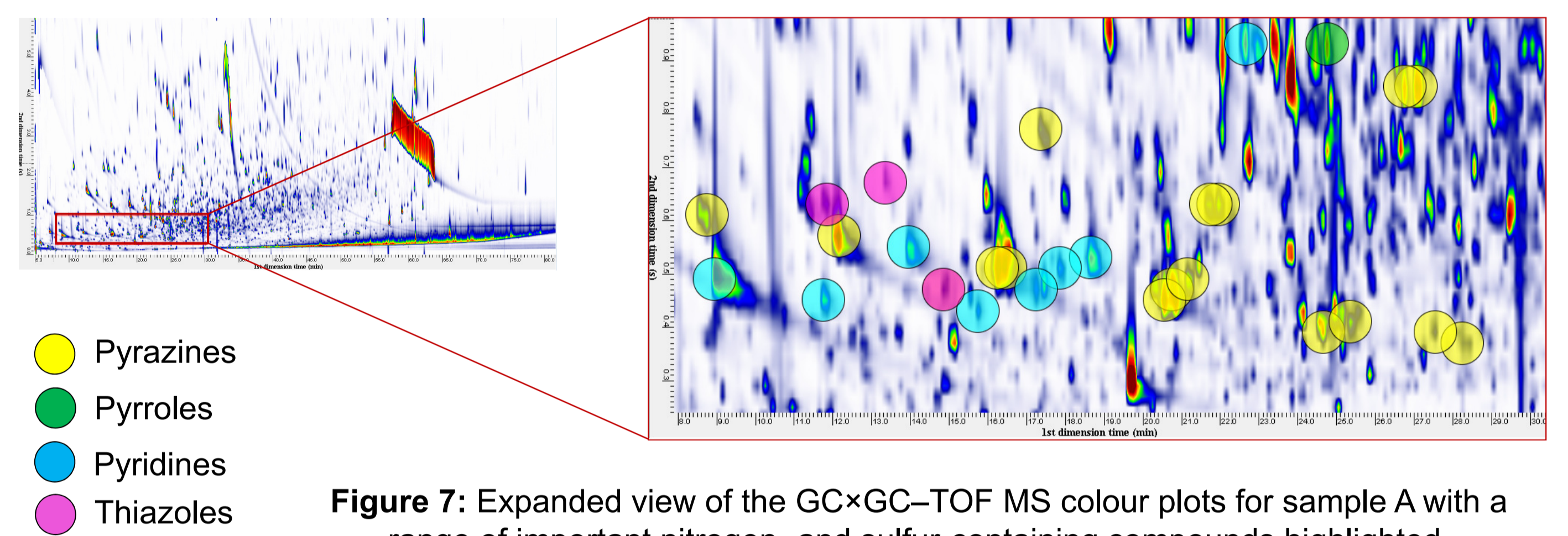


Figure 7: Expanded view of the GC×GC–TOF MS colour plots for sample A with a range of important nitrogen- and sulfur-containing compounds highlighted.

The script shown in Figure 8 was used to identify 2-acetyl-3-methylpyrazine within the complex samples, overcoming the 'needle in a haystack' problem. This compound is a key contributor to a hazelnut taste/aroma in coffee.

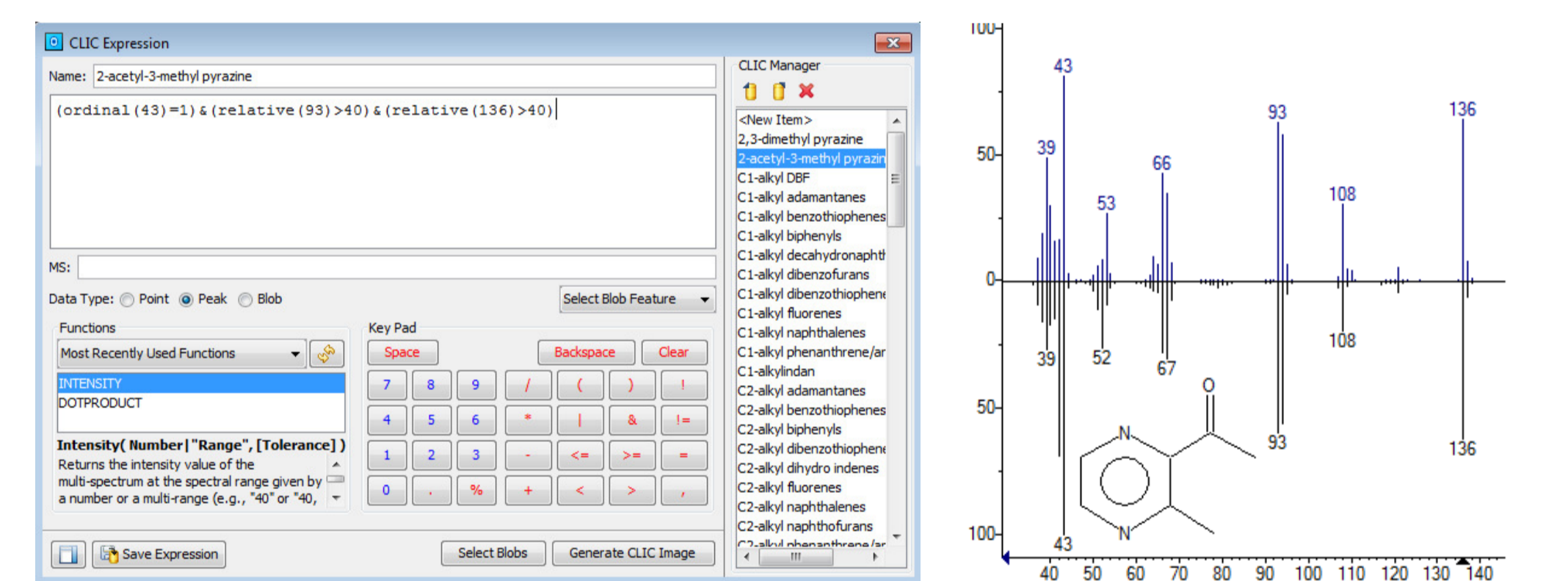


Figure 8: A simple scripting function using mass spectral qualifiers for the identification of 2-acetyl-3-methylpyrazine (left), and the NIST match for the peak found in sample A (right).

Applying the template to all three samples (Figure 9) showed that sample C had a very low abundance of pyrazines compared with samples A and B. Pyrazines generally impart a pleasant nutty/earthy/roasted aroma to coffee, and in certain cases can have extremely low odour thresholds (<0.1 ppb in the case of 2,3-diethyl-5-methylpyrazine).

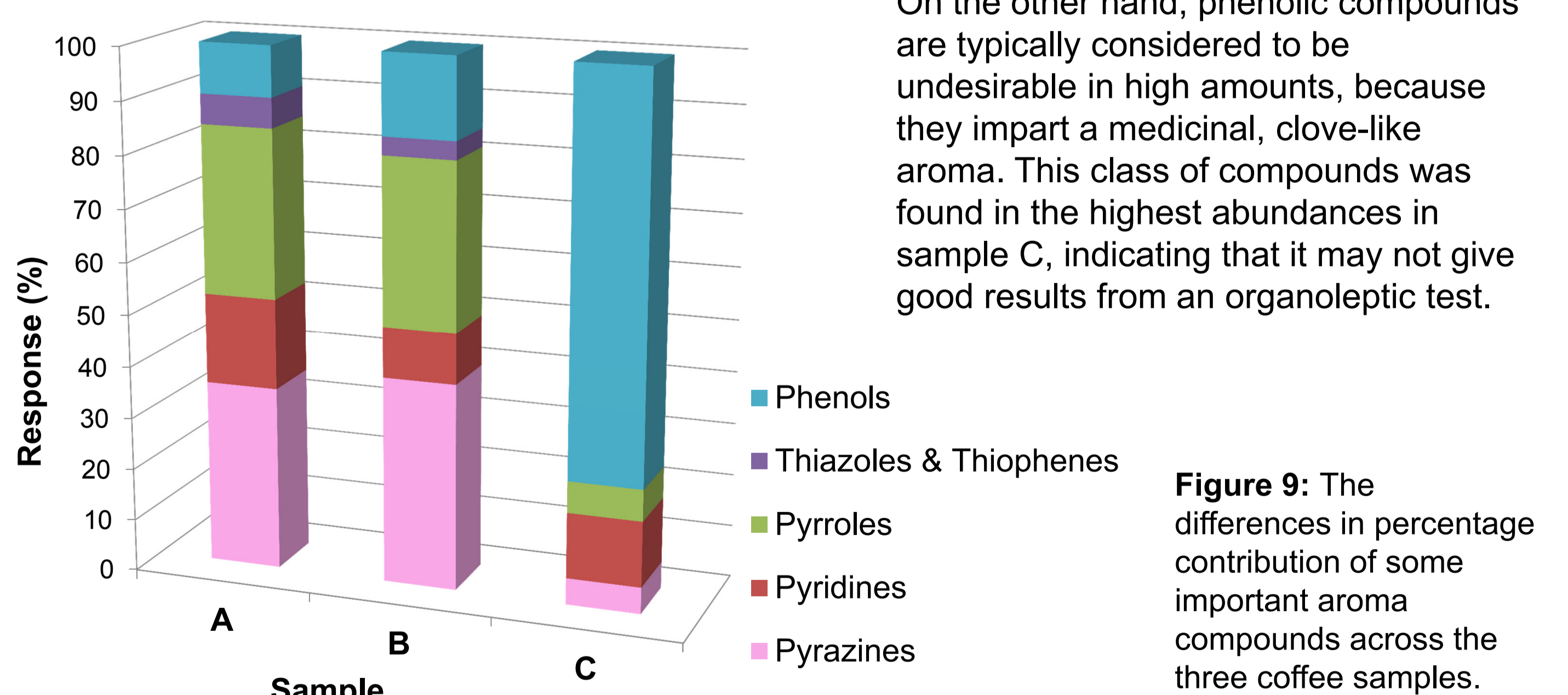


Figure 9: The differences in percentage contribution of some important aroma compounds across the three coffee samples.

Conclusions

This work has shown that GC×GC–TOF MS, using a BenchTOF-HD as the detector, is ideally suited to the analysis of complex beverage samples, due to its combination of high sensitivity, spectral quality and chromatographic resolving power.

The excellent spectral quality provides confident identification of both targets and unknowns, even at trace levels. Thus, the system provides enhanced confidence when monitoring product quality in the food industry.

GC×GC–TOF MS was able to identify a range of minor constituents in the coffee samples that may have escaped notice on a conventional GC–MS system due to numerous coelutions with higher-loading peaks.