Technical Overview

QuickProbe Dual Configurations for Forensic Workflows

Providing flexibility and robustness on a single GC/MS system

Authors

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Abstract

Analysis of unknown powders, tablets, and liquids by forensic drug chemists have routinely used capillary chromatography with mass selective detectors (MSD). However, this usually requires sample preparation and/or acid/base extraction, and consistently includes runtimes from 10 to 30 minutes for general sample screenings.[1] Agilent QuickProbe GC/MS analysis can produce "classical EI" spectral identification of compounds in a variety of sample matrices with minimal to no sample preparation in approximately a minute. The purpose of this research is to demonstrate the ability of the QuickProbe GC/MS system to be successfully incorporated into the current forensic workflow on existing GC/MS systems.[2,3] This work outlines in detail how the technique can be configured on an existing Agilent 5977/8890 or Agilent 7890 GC in three separate configurations, while maintaining the current conventional capillary GC/MS capabilities. QuickProbe is controlled by Agilent MassHunter software, with access to Acquisition control, Qualitative Analysis, Unknowns Analysis integration using deconvolution, and reporting templates.

Introduction

The QuickProbe GC/MS system (QP) comes with its own heated atmospheric capillary split/splitless inlet and a short (approximately 2 m) high-temperature analytical column. The factory configuration QP column is approximately 1.5 m long with a 0.25 mm id, connected to an Agilent Ultimate Union. The outlet of the Ultimate Union is attached to a 60 to 70 cm-long restrictor column, 0.18 mm id, with 0.18 µm film sealed to the MSD transfer line and running into the ion source (Figure 2). This arrangement reduces flow rate into the ion source by a factor of approximately two, and can be varied by adjusting the restrictor length. Typical flows are approximately 1.5 to 2.4 mL/min into the ion source (2.4 mL/min at 60 °C initial column temperature) and approximately 20 mL/min into the split vents. This results in a total QuickProbe injector liner flow rate of ~22 mL/min, with a split ratio of approximately 10.[4] Agilent recommends a DB-1ht column rated at 400 °C for both columns. The factory default column is an Agilent J&W DB-1ht, high-temperature GC column, 2 m × 0.25 mm, 0.1 µm (G3903-61006). High-temperature columns are recommended with the QP due to its fast-ramping capabilities of 16 °C per second (960 °C per minute). However, column stationary phase chemistry can be customized based on the individual application requirements. DB-1ms and DB-5ms are common universal phase chemistries used in forensic screening and analysis, and they are considered standard and semistandard nonpolar compound separation columns, respectively.[5,6] A narrower QP analytical column was used in this study to help reduce flow into the MSD and generate better peak resolution. Data generated in all three dual-column configurations used an approximately 2 m column length, cut from an Agilent J&W VF-5ms column, 12 m × 0.20 mm, 0.33 µm (CP98935) as the QP analytical column. The conventional GC analytical column installed in the Agilent 8890 GC split/splitless inlet in this study used an Agilent J&W DB-5ms EVDX 25 m × 0.20 mm, 0.33 µm (128-8522) or an Agilent DB-5ms 20 m × 0.18 mm, 0.18 µm column (121-5522UI) to generate conventional GC/MS analyte chromatographic data for confirmational purposes, allowing for a single system with a dual configuration to screen and identify sample analytes.

Agilent recommends the use of a 6 mm drawout or extractor lens, depending on the version of source, to be installed in the ionization source. This helps to reduce the possibility of overloading the detector signal. The application was tested with both 3 and 6 mm extractor lenses, and both performed well. The data in this work were all generated using the 6 mm extractor lens. The key factors that determine which lens to use depend on sampling technique, concentration, and physical properties of the analytes of interest. Analytes can be found in Table 1.

Table 1. Application-specific examples of recommended drawout and extractor lens diameters.

Analyte Concentration and Matrix	Drawout or Extractor Diameter
Explosive Residue Soil Extract	3 mm lens
Illicit Forensic Street Drugs	6 mm lens
Organic Gunshot Residue	3 mm lens
Plastic Polymer Impurities	6 mm lens

A separate GC/MS-QP method parameter screen exists when using the QP, which contains the QP temperature ramp, inlet temperature, split mode, and inlet pressure settings for acquisition. During GC/MS-QP acquisition, the MS parameter screen is set to typical GC/MS scan acquisition parameters, while the GC parameter screens are used to set an isothermal oven temperature appropriate for the analysis. This controls the GC inlet temperature, flow settings for the GC analytical column, and MSD transfer line temperature while the QP is in acquisition mode. When acquiring conventional GC/MS data, the QP is set to Standby mode, reducing the QP inlet head pressure. In this mode, the standard GC and MS parameter windows are applied to generate customary conventional GC/MS capillary data. All three configurations manifest within the GC oven and on the MSD interface/transfer line. Figure 1 depicts the external view of an Agilent 5977B/8890 GC/MS-QP system with the atmospheric split/splitless inlet on the right side of the QP module. Figure 2 illustrates the overall flow path and sample introduction on the factory standalone GC/MS-QP system. Figure 3 explains the dual analytical configuration of using a GC inlet and modifications to the MSD transfer line interface for dual GC/MS and GC/MS-QP analysis.

Figure 1. Agilent 5977B GC/MSD with an Agilent 8890 GC system and Agilent QuickProbe GC/MS system.

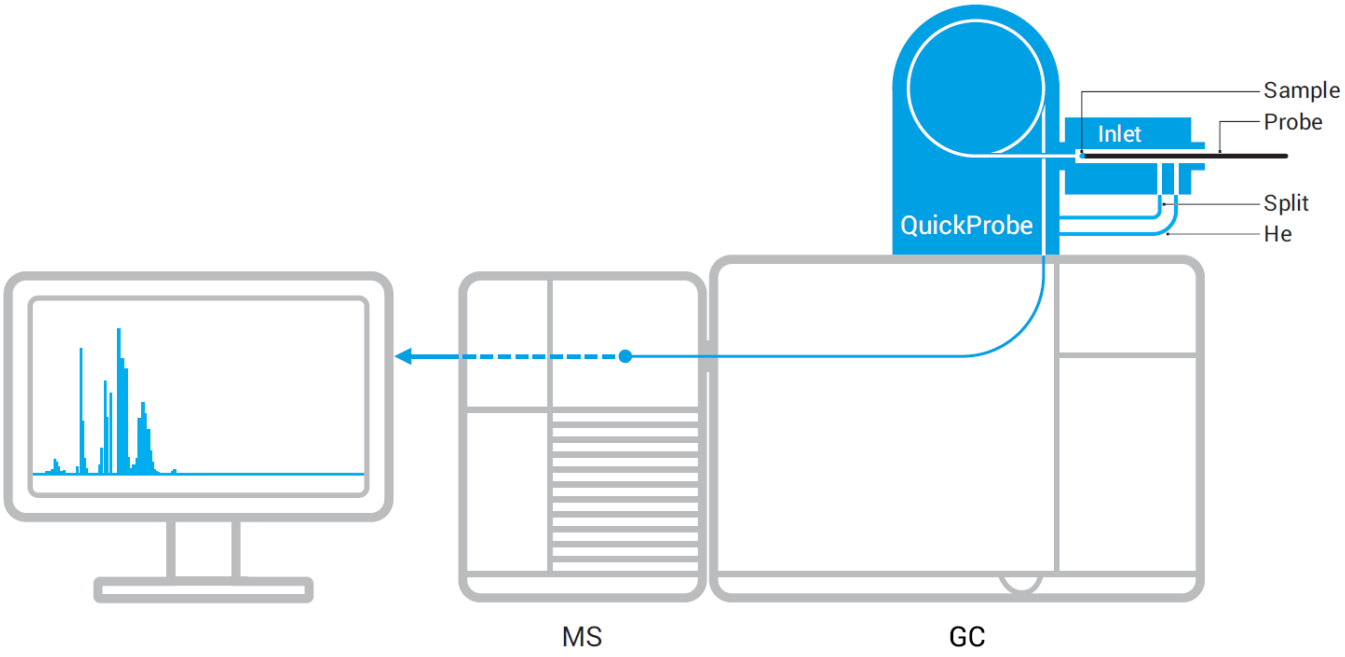


Figure 2. Standalone Agilent QuickProbe GC/MS flow and sample introduction pathway.

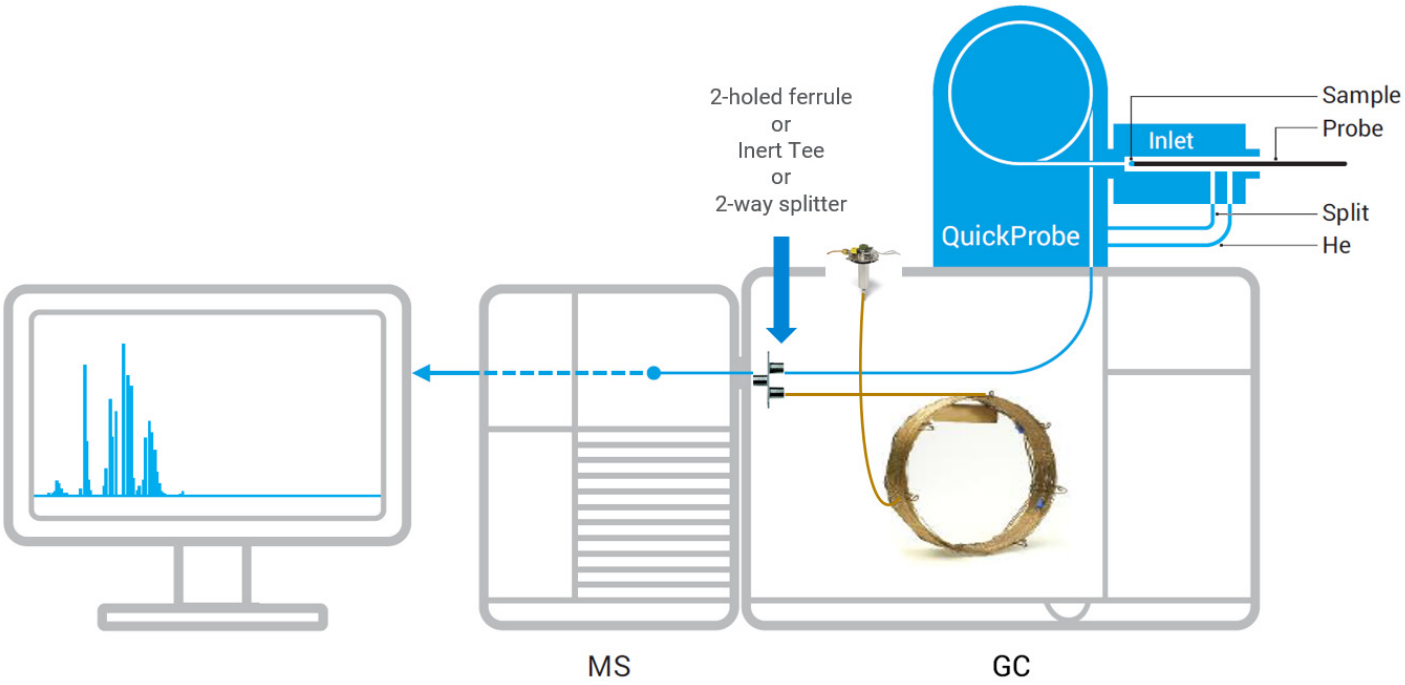


Figure 3. Dual-configuration GC/MS and GC/MS-QP flow, and sample introduction pathway with dual analytical columns.

Experimental

Materials and consumables are grouped accordingly with the dual configurations of 1, 2, and 3.

Configuration 1

- Agilent 2-holed ferrule, 0.4 mm id, 15% graphite/85% Vespel, 0.1 to 0.25 mm column, 10/pk (part number 5062-3580)
- Agilent column nut for MS interface (part number 05988-20066)
- Agilent column installation tool 5973 series, 5975 A/B/C/C TAD/E, 5977 series, and 7000 (G1099-20030)

Configuration 2

- Agilent Tee union, inert, for capillary flow technology (G3184-60065)
- Agilent QuickSwap Transfer Line Locking Nut (G3185-20501)
- Agilent Flexible Metal ferrule, gold-plated, 0.4 mm id, for 0.1 to 0.25 mm column, 10/pk (G2855-28501)
- Agilent Ultimate Plus deactivated fused silica tubing, 5 m, 0.18 mm uncoated (CP801805)

Configuration 3

- Agilent Compact 2-way splitter, inert, without makeup gas (G3181-60500)
- Agilent Flexible Metal ferrule, gold-plated, 0.4 mm id, for 0.1 to 0.25 mm column, 10/pk (G2855-28501)
- Agilent Ultimate Plus deactivated fused silica tubing, 5 m, 0.18 mm uncoated (CP801805)
- Agilent Ferrule, 0.4 mm id, 15% graphite/85% Vespel 0.1 to 0.25 mm column, 10/pk (part number 5181-3323)

Common consumables for all configurations used in this study:

- Agilent GC analytical column: J&W DB-5ms Ultra Inert GC Column, 20 m × 0.18 mm, 0.18 µm (121-5522UI)
- Agilent GC analytical column: J&W DB-5ms EVDX GC Column, 25 m × 0.20 mm, 0.33 µm (128-8522)
- Agilent QuickProbe analytical column: J&W VF-5ms GC Column, 12 m × 0.20 mm, 0.33 µm (CP8935)
- Agilent GC liner: Splitless, UI, Fritted Liner, Low, 870 µL, 4 mm, 5/pk (part number 5190-5112-005)
- Agilent QuickProbe liner: Ultra Inert, 4 mm id, fritted bottom (part number 5190-5104)
- Agilent QuickProbe probe, holder (G3971-60200)
- Agilent QuickProbe probe, round tip, 100/pk (part number 5190-5118)

Configuration 1: 2-Holed ferrule

Simple and cost-effective, but least flexible, since column changes require venting the MS

The first configuration is the simplest and most cost-effective, but has the least flexibility when the need arises to vent the MS to change the QP or GC analytical columns. This configuration uses the older style brass nut (part number 05988-20066) to accommodate the larger 2-holed Vespel/graphite ferrule (part number 5062-3580). Figure 4 illustrates the QP and conventional analytical columns threaded through the brass nut and 2-holed ferrule with a blue septum (usually found on new/stored Agilent GC columns) to keep the columns in position and from twisting around each other. **Note:** The 2-holed ferrule still goes in backwards with the flat side facing/flush against the MSD transfer line to allow for a proper vacuum seal. The next step in Figure 5 is optional, but does make the positioning process of both columns in the MS transfer line easier and more straightforward. Figure 5 uses a column insertion tool (G1099-20030) for accurate placement of both columns in the MS transfer line, allowing both columns to be locked into place, minimizing column movement during installation.

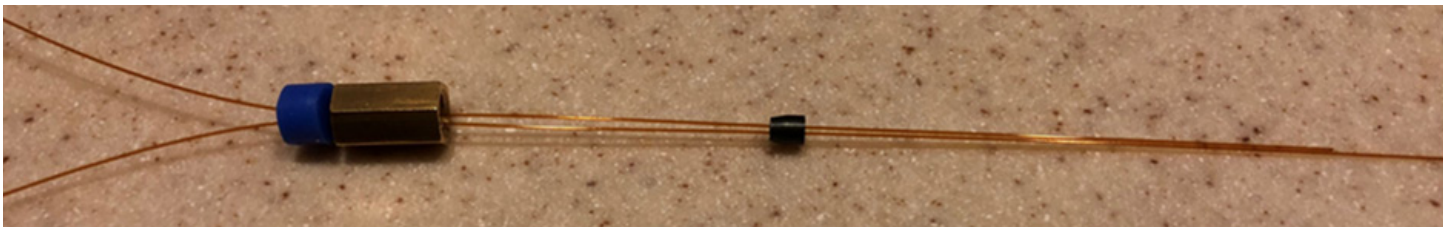


Figure 4. Column placement exaggerated to illustrate the proper use of the tool.

Figure 5. Column placement exaggerated to illustrate the proper use of the column insertion tool.

Configuration 2: Inert tee union

Flexible for column changes without venting the MS, but requires accurate transfer line length and placement

The second configuration brings both the QP and GC analytical columns into the MS transfer line via an inert tee union (G3184-60065). In this configuration, vacuum-tight connections are made using flexible gold metal ferrules (G2855-28501) on both column connections into the inert tee and on the MS transfer line column that enters on the backside of the inert tee union (Figure 6). In this study, the MSD transfer line consists of an Ultimate Plus fused silica deactivated piece of tubing 0.18 mm × ~17 cm in length. The MS transfer line is cut to approximately 17 cm to allow for the proper placement into the MS source, and uses a section cut from a 5 meter spool of tubing (CP801805). Figure 7 displays the installation of both the QP and GC analytical columns into the inert tee. This configuration provides more flexibility for changing either analytical column easily and/or capping off the tee inlet using only one of the analytical techniques.

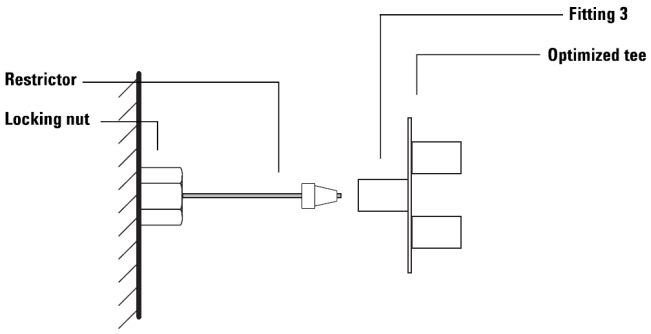


Figure 6. Installing the transfer line restrictor and inert tee union.

Configuration 3: 2-way splitter without makeup gas

Easy column and transfer line installation, flexible column changes without venting the MS, and the most robust choice

The third and last configuration discussed is the author's recommended configuration, due to its ease-of-access for column installation, troubleshooting, and overall robustness of the configuration. This configuration allows for quick column changes without the need for venting the MS system. However, it is recommended that the source and quadrupole temperature should be less than 100 °C, and that new ferrule-to-column connections have already been made prior to changing either column. Using the 2-way splitter option (G3181B, G3181-60500) places all column connections in front of the oven and, in this example, mounted on the oven wall close to the MS transfer line. Depicted in Figures 8 and 9, the bottom position of the 2-way splitter is occupied by the GC analytical column, which has its flow sweeping the next port (middle position) with the QP analytical column, followed by the MS transfer line interface column (CP801805) coming out of the top position and into the MS transfer line interface. The MS transfer line uses the same Ultimate Plus deactivated fused silica (CP801805) as configuration 2, and is cut to the proper length (approximately 25 to 30 cm) for ease of correct placement into the MS transfer line. All three splitter connections use gold ferrules (G2855-28501, also used in configuration 2). The MS interface uses the standard Vespel/graphite ferrule (5181-3323), 0.4 mm id, used in the GC or QP inlet. The shorter single-holed ferrule still goes in backwards with the flat side facing/flush against the MSD transfer line to allow for a proper vacuum seal.

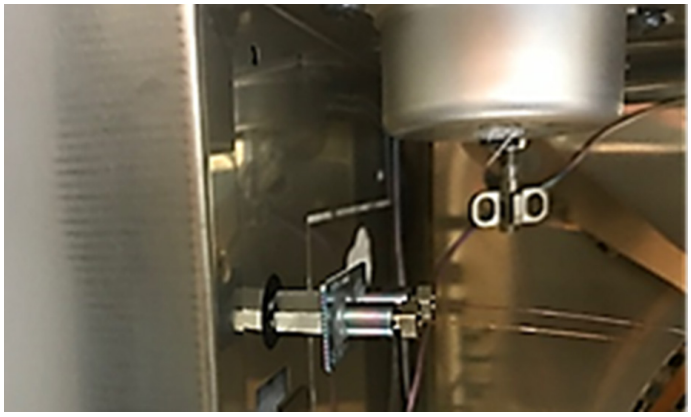


Figure 7. Configuration 2: Inert tee installed on the MS transfer line with the QP and GC analytical columns.

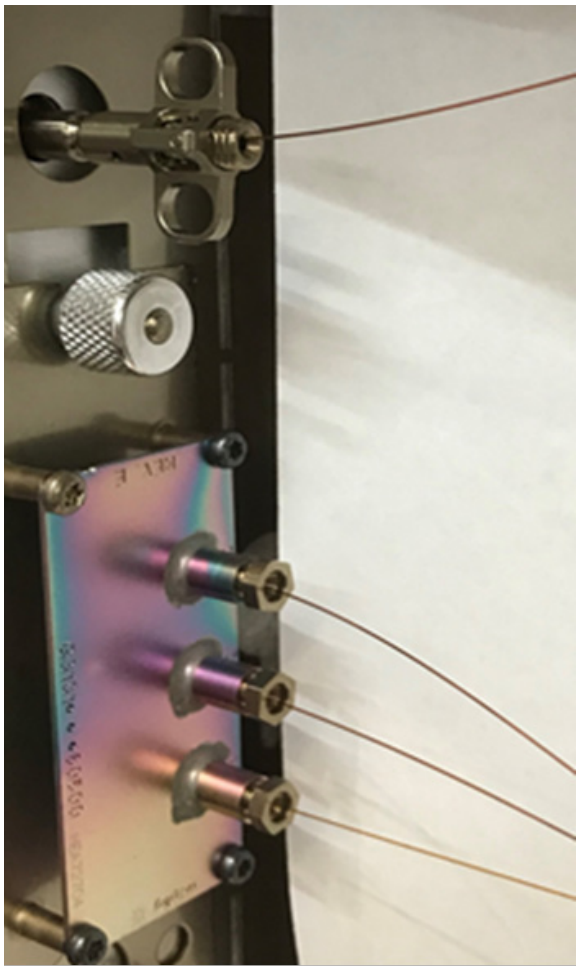


Figure 8. Configuration 3 displays the 2-way splitter installed on the inside of the GC oven. The bottom inlet is the GC analytical column. The middle inlet is the QP analytical column, and the top inlet is the MS transfer line.

Results and discussion

All three configurations were run separately, acquiring QP data first (screening) and then generating conventional GC/MS data as a confirmatory data point, all using an autotune (atune.u) with a 285 °C source temperature. The optimal source temperature was determined by running various classes of drugs (phenylethylamines, cocaine, benzodiazepines, opiates, fentanyl analogs) with the source at 325, 285, and 230 °C, and comparing peak shape and overall responses of the drugs at the three different source temperatures. Tables 2 and 3 illustrate the typical parameters used, based on a GC/MS conventional analysis and on a GC/MS-QP analysis, respectively. There were slight differences in GC oven ramps based on the GC/MS analytical column used, along with slight variations in QP ramping to optimize separation and speed of analysis during the process of this study. All samples were collected on separate round-tipped QP borosilicate probes. All probes were prewashed in hexane, sonicated for five minutes, allowed to dry, and stored in the GC oven until used in analysis.

Various types of sample matrices were explored and successfully run under all three configurations. Figure 9 shows an example of data acquired under configuration 1 (2-holed ferrule). The top total ion chromatogram (TIC) consists of a methanolic dilution of a 5 mg oxycodone tablet scraping on a 20 m DB-5ms column with an 18-minute runtime, while the bottom (TIC) represents QP analysis of a tablet scraping from the same tablet (tablet surface scratched with a QP round-tip probe) on an approximately 2-meter VF-5ms column requiring a one-minute runtime for analytes to achieve separation. Once the initial tablet scraping was analyzed on the GC/MS-QP, additional tablet powder was produced (approximately 0.5 mg), and added to a 2 mL sample vial. Approximately 1.5 to 2 mL methanol was added to the vial and placed in the ALS tray for GC/MS conventional analysis and confirmation.

Figure 10 was collected under configuration 2 (inert tee union) and comprises an unextracted mixture of 15% lemon/orange oil concentrate that was spiked with 50 ppm cannabidiol, 100 ppm of delta-9-tetrahydrocannabinol, and cannabinol. The tip of a round probe was dipped approximately 3 mm into the unextracted oil solution and injected into the QP inlet for a 1.5-minute runtime. The remainder of the solution was transferred to a 2 mL vial and placed in the ALS tray for confirmation during a 25 minute run.

Data from the fentanyl analog mixture is shown in Figure 11 using configuration 3 (2-way splitter) and depicts a standard of 11 fentanyl analogs at 35 ppm in methanol, run on a 25 m GC/MS analytical column (top) and the 2 m GC/MS-QP column (bottom). Note the 25 m, 25-minute method allowed for almost baseline separation between methacryl fentanyl, 3-methyl fentanyl, and alpha-methyl fentanyl, and did produce baseline resolution of cyclopropyl fentanyl and crotonyl fentanyl. In the QP data, methacryl-, 3-methyl-, and alpha-methyl fentanyl generated one large Gaussian peak, as did the cyclopropyl and crotonyl fentanyl. However, utilizing the deconvolution software in Agilent MassHunter Unknowns Analysis, all three coeluting fentanyl analogs were identified, as seen in Figure 12. Cyclopropyl and crotonyl fentanyl are structural isomers, and these two compounds are difficult to identify owing to their similarity in structure, chromatographic behavior, and nearly identical mass spectrum.[7] Due to the spectral similarities, the deconvolution software cannot identify the spectral differences between cyclopropyl and crotonyl fentanyl when coeluting (Figure 13).

Elution order: Acetyl norfentanyl, norfentanyl, butyl norfentanyl, 4-ANPP, fentanyl carbamate, fentanyl, methacryl fentanyl, 3-methyl fentanyl, alpha-methyl fentanyl, cyclopropyl fentanyl, and crotonyl fentanyl.

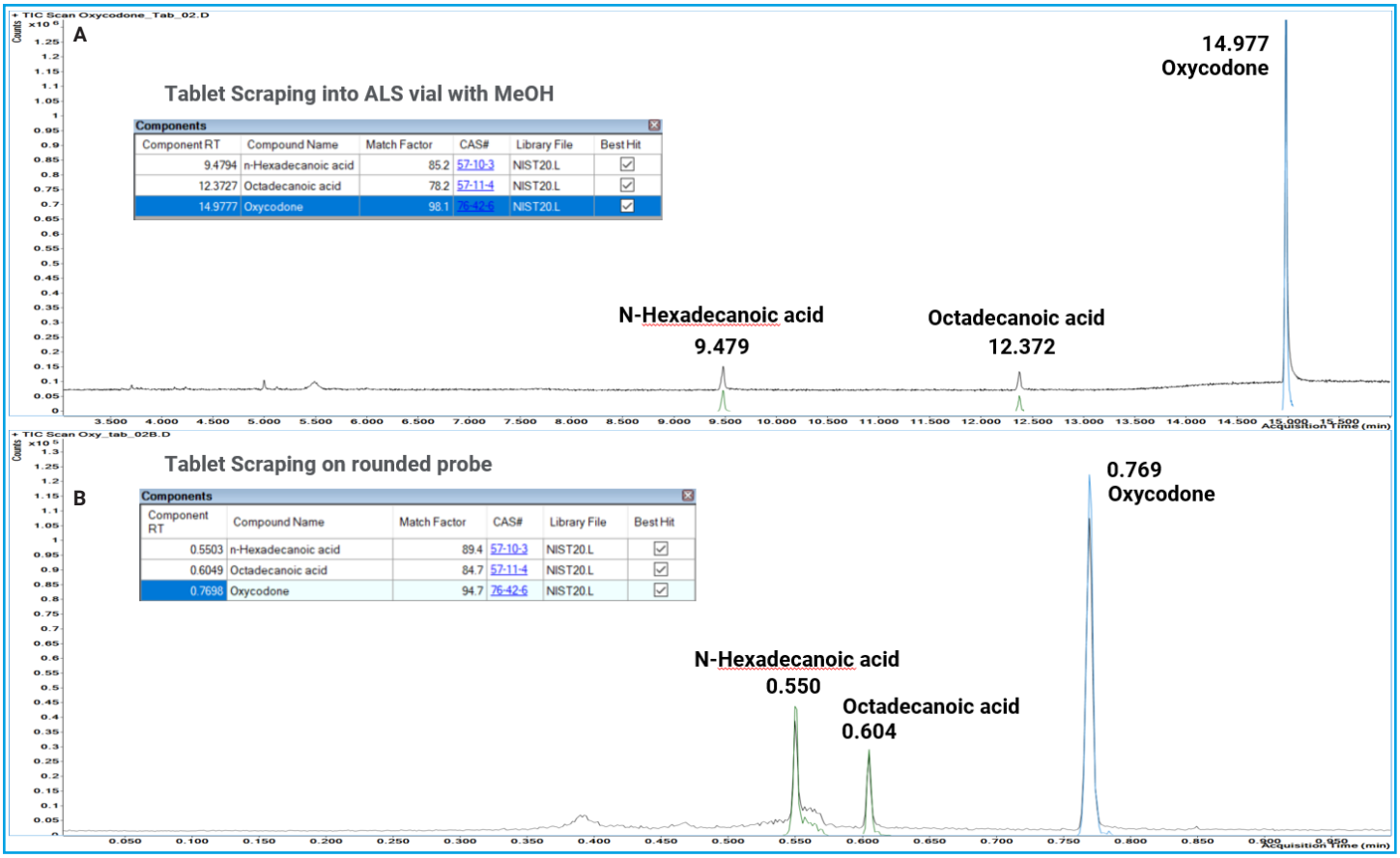


Figure 9. (A) GC/MS conventional 17-minute run of tablet scraping on a 20 m × 0.18 mm, 0.18 µm Agilent DB-5ms column; (B) QP 1-minute screening run of tablet scraping on a 2 m × 0.20 mm, 0.33 µm Agilent VF-5ms column.

Table 2. Typical instrument parameters for GC/MS conventional analytical data acquisition for fentanyl analog mix in lemon/orange oil matrix.

Parameter	Setting
Injection Source	Autosampler tower/tray
Injection Volume	1 µL
GC Split/Splitless Inlet	250 °C, Split mode 10:1
QP Split/Splitless Inlet	250 °C, Capped off
GC Analytical Column Temperature Program	215 °C (hold 0 mins) 5 °C/min to 315 °C (hold 5 mins) 25-minute runtime
QP Analytical Column Temperature Program	Stand-By Mode Isothermal at 40 °C Helium at 5 psi
GC Analytical Column Flow	Helium; 1.0 mL/min
MS Transfer Line Temperature	285 °C
Ion Source Temperature	285 °C; atune_6mm_285.u
Quadrupole Temperature	150 °C
Scan Range	40 to 550 *m/z*
Gain	1
Threshold	0
A/D Samples	2

Table 3. Typical instrument parameters for GC/MS-QP screening data acquisition for fentanyl analog mix in lemon/orange oil matrix.

Parameter	Setting
Injection Source	Manual QP sample holder
Injection Volume	Solid particles or dip tip ~3 mm
GC Split/Splitless Inlet	250 °C, Split mode 10:1
QP Split/Splitless Inlet	250 °C, Split mode 10:1
GC Analytical Column Temperature Program	Isothermal at 285 °C
QP Analytical Column Temperature Program	60 °C (hold 0 sec) 7 °C/s to 225 °C (hold 5 sec) 12 °C/s to 315 °C (hold 45 sec) Helium at 15 psi 1.3-minute runtime
GC Analytical Column Flow	Helium; 1.0 mL/min
MS Transfer Line Temperature	285 °C
Ion Source Temperature	285 °C; atune_6mm_285.u
Quadrupole Temperature	150 °C
Scan Range	40 to 550 *m/z*
Gain	1
Threshold	0
A/D Samples	2

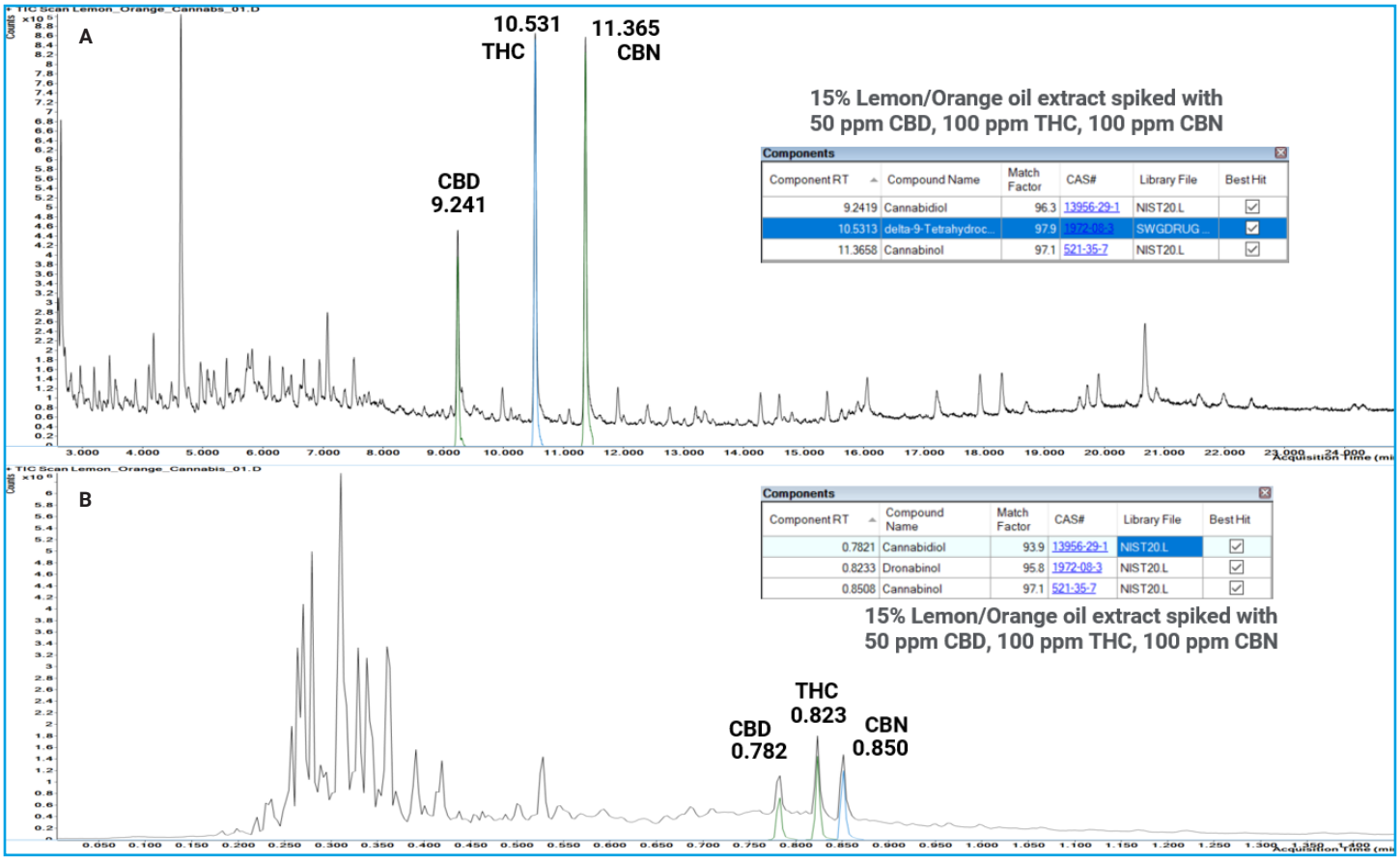


Figure 10. (A) GC/MS conventional 25-minute run of fentanyl analogs on an Agilent J&W DB-5ms EVDX column, 25 m × 0.20 mm, 0.33 µm. (B) QP 1.5-minute screening run of the same fentanyl analogs on an Agilent J&W VF-5ms column, 2 m × 0.20 mm, 0.33 µm.

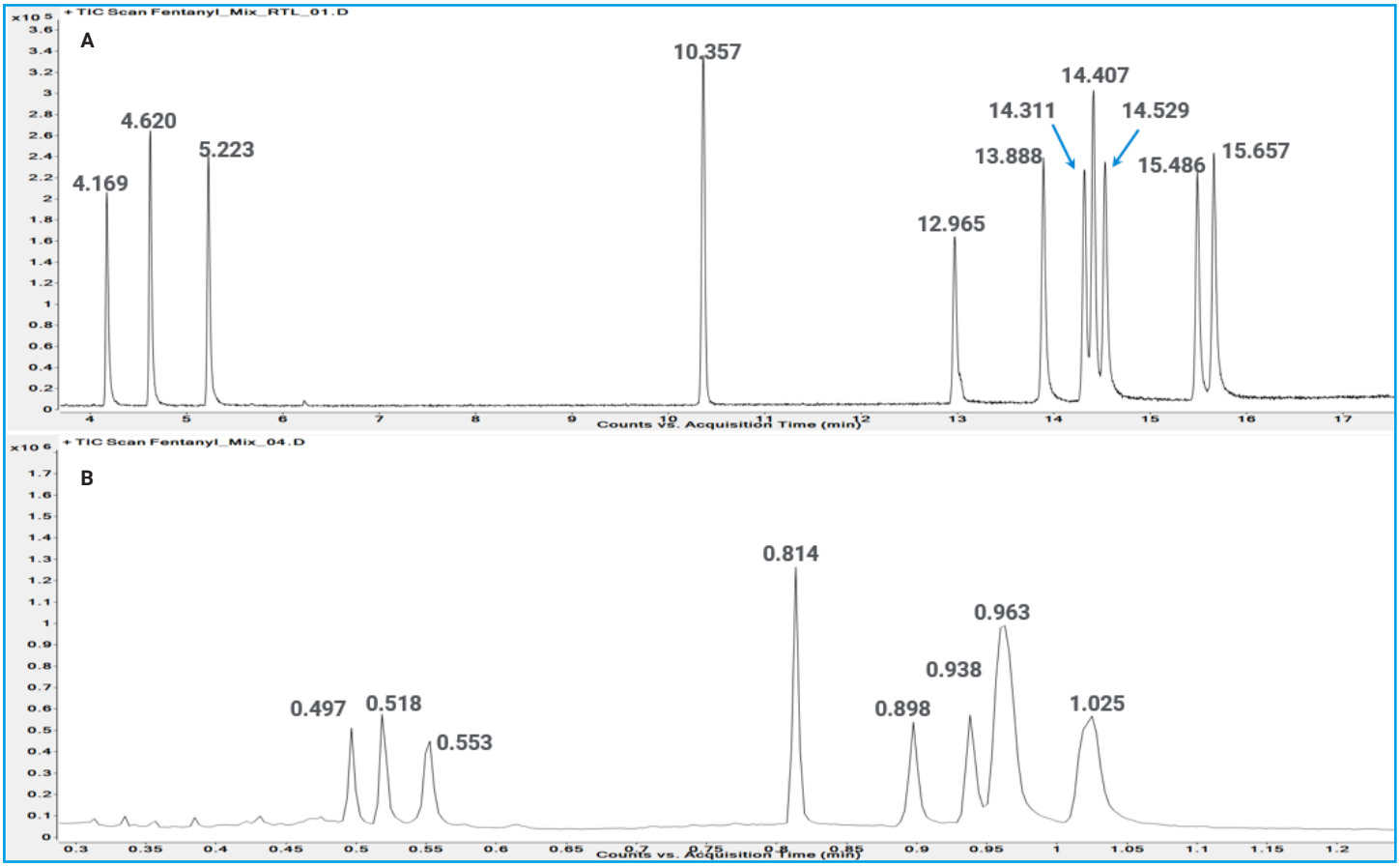
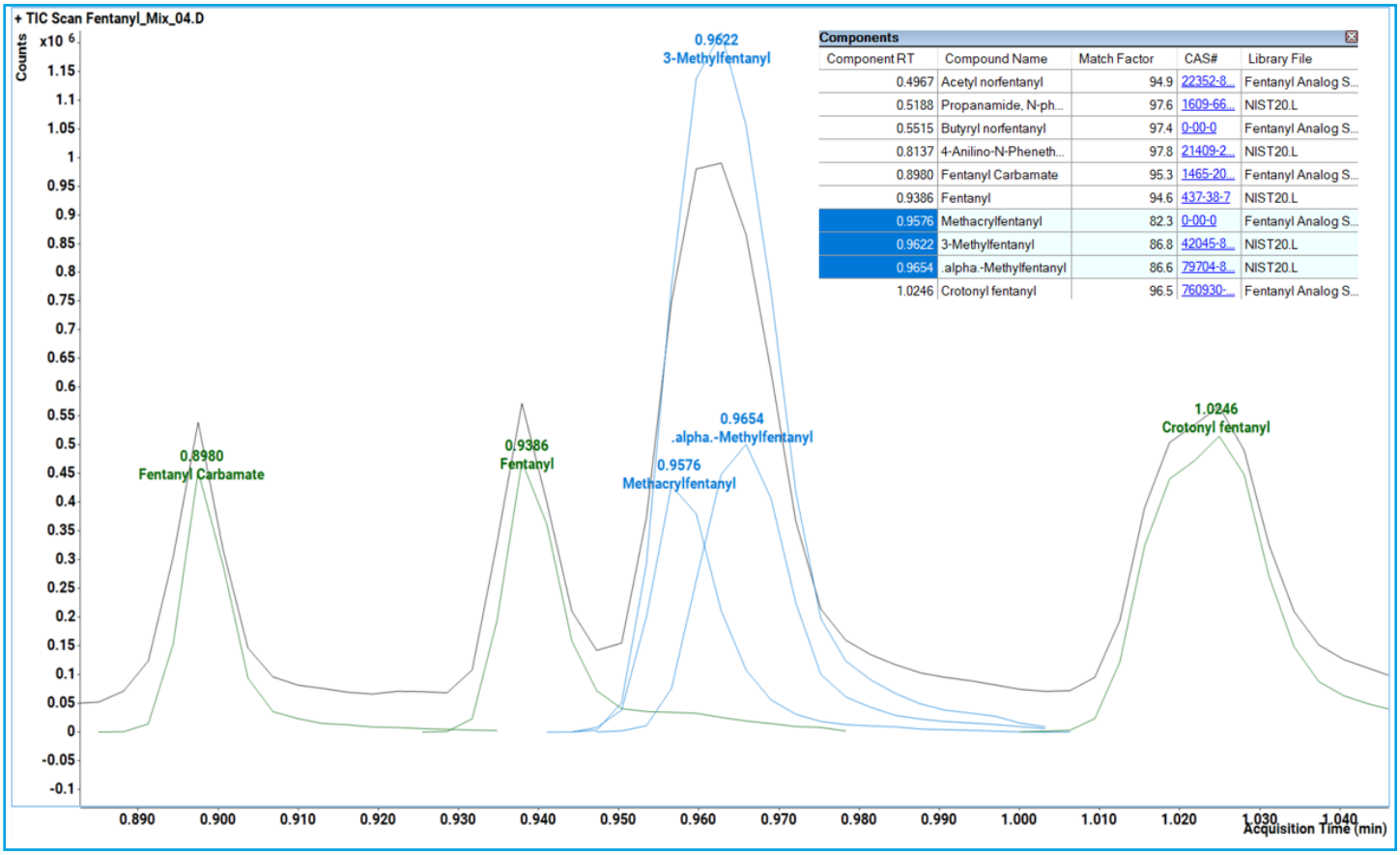


Figure 11. (A) GC/MS conventional 25-minute run of fentanyl analogs on an Agilent J&W DB-5ms EVDX column, 25 m × 0.20 mm, 0.33 µm. (B) QP 1-minute screening run of the same fentanyl analogs on an Agilent J&W VF-5ms column, 2 m × 0.20 mm, 0.33 µm.



Components

Component RT	Compound Name	Match Factor	CAS#	Library File
0.4967	Acetyl norfentanyl	94.9	22352-8...	Fentanyl Analog S...
0.5188	Propanamide, N-ph...	97.6	1609-66...	NIST20.L
0.5515	Butyryl norfentanyl	97.4	0-00-0	Fentanyl Analog S...
0.8137	4-Anilino-N-Pheneth...	97.8	21409-2...	NIST20.L
0.8980	Fentanyl Carbamate	95.3	1465-20...	Fentanyl Analog S...
0.9386	Fentanyl	94.6	437-38-7	NIST20.L
0.9576	Methacrylfentanyl	82.3	0-00-0	Fentanyl Analog S...
0.9622	3-Methylfentanyl	86.8	42045-8...	NIST20.L
0.9654	.alpha.-Methylfentanyl	86.6	79704-8...	NIST20.L
1.0246	Crotonyl fentanyl	96.5	760930-...	Fentanyl Analog S...

Figure 12. Coelution of methacryl-, 3-methyl-, and alpha-methyl fentanyl, and subsequent identification using deconvolution software.

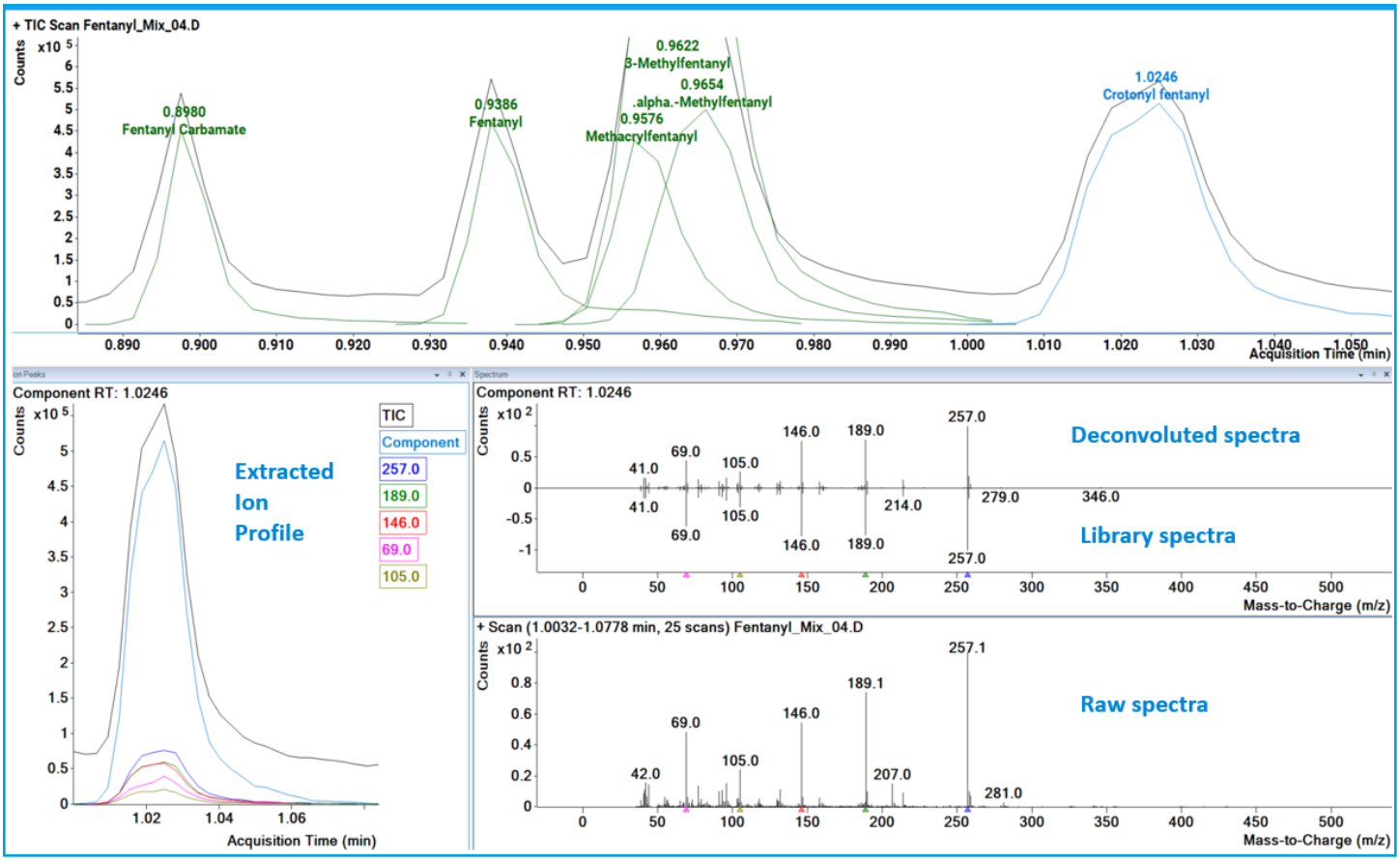


Figure 13. Coelution of cyclopropyl and crotonyl fentanyl, and mass spectral similarities.

Conclusion

All three configurations allow the use of a new or existing GC/MS system for screening and confirmations on a single system. The 2-holed ferrule configuration is the most economical, and in principle, the easiest to configure; however, it does take some technique to place both the GC analytical column and the QP analytical in the correct transfer line depth without twisting and possibly breaking one or both columns. This configuration also requires venting of the MS to change either column. The inert tee provides a mechanism to change columns quickly, but venting is still necessary. The inert tee does require gold ferrules and a special swaging tool for connections, and an accurate measurement and cut for the MS transfer line. The third and final 2-way splitter configuration allows for vent-free column changes (cooled source and quadrupole) and necessitates the use of the gold ferrules and swaging tool. The 2-way splitter is also the most expensive configuration, but is the most rugged in the long-term, and is the configuration the author recommends. Choosing any of these configurations in conjunction with the Agilent QuickProbe GC/MS system increases screening efficiency with minimal to no sample preparation, generates "classical EI" spectra for library matching, and offers an approximately one-minute runtime, while using conventional conformational methods on an existing GC/MS system.

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RA45132.467025463

Printed in the USA, November 13, 2023
5994-6889EN

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