K46 Emergence of Delta-8 Tetrahydrocannabinol (THC) in Driving Under the Influence of Drugs (DUID) Investigation Casework

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Learning Overview: After attending this presentation, attendees will be able to identify delta-8 THC and its appearance in various marijuana-containing products. Additionally, this presentation will illustrate an important analytical challenge related to constantly changing drug trends and how it was addressed in a high-throughput laboratory setting.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by providing information on the recent increase in prevalence of THC isomers in DUID cases. Furthermore, an example method is described, along with its validation and application, showing how it effectively reduced both repeat testing and cancellations due to unresolved peaks.

Cannabinoids are the most frequently reported illicit drug class in DUID investigation casework, with a consistent positivity rate over 50%. Since September of last year, the cancellation rate for the cannabinoids confirmation performed using a 2D High-Performance Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) demonstrated a steady increase (from 1.3% to 3.8%) due to the presence of unresolved interfering substances. The observed interference demonstrates a distinctive pattern affecting identifications and quantification of both delta-9 THC and delta-9 Carboxy THC (THCCOOH).

As a part of an investigation, an e-cigarette device containing an amber-colored viscous solution was submitted to the laboratory. The oily substance was diluted 100-fold for analysis. Two peaks with equally high response were identified as delta-9 THC and delta-8 THC, in addition to a presence of Cannabidiol (CBD) and four other unidentified components in the e-cigarette cartridge. Ultimately, additional work revealed delta-8 THC (*RT*=5.01min) and delta-8 THCCOOH (*RT*=3.05min) to be responsible for the observed interferences with delta-9 THC (*RT*=4.92min) and delta-9 THCCOOH (*RT*=3.14min), respectively.

Delta-9 THC and delta-8 THC are constitutional isomers ($C_{21}H_{30}O_2$; MW 314.46), as are their carboxy metabolites ($C_{21}H_{28}O_4$; MW 344.44). The similarity between the two isomers was not previously a challenge due to the relatively small concentrations of delta-8 THC encountered after traditional marijuana consumption. Delta-8 THC is a naturally occurring cannabinoid found in most cannabis plants. It is typically present at low concentrations (less than 1% of dried cannabis flowers). It is psychoactive, but has a lower potency as compared to delta-9 THC. Recently, delta-8 THC has been marketed as an alternative to marijuana. Delta-8 THC has been identified in botanicals, e-liquids intended for vaping, and edibles. The increase in cancellations due to delta-8 compounds required modification of the existing method that had been successfully employed for the past three-and-a-half years.

To separate the delta-8 and delta-9 isomers, a new analytical method was developed and validated. Cannabinoids were extracted from whole blood using liquid-liquid extraction, separated in a 2D LC system with a Phenomenex® SecurityGuard™ Cartridge (C6-Phenyl, 4 X 2mm) as a trap column and a Phenomenex® Kinetex® C18 (100 x 3mm; 2.6micron) as an analytical column, over a runtime of 11.5min and detected by an AB SCIEX™ API 4500 system equipped with an Electrospray Ionization (ESI) source operating in positive ionization mode with scheduled multiple reaction mass spectrometric monitoring. The method validation protocol was based on the Scientific Working Group for Forensic Toxicology (SWGTOX) guidelines, including linearity, limit of detection, lower limit of quantitation, precision and accuracy, interfering substances, matrix effect, dilution of samples, and carryover. The calibration for THC and THCCOOH was linear from 0.5 to 50ng/mL and 5.0 to 500ng/mL, respectively. The minimum total precision (described as %CV) and accuracy (shown in %difference) were 3.4% and -3.4% for THC and 3.1% and -3.9% for THCCOOH, respectively. The method met validation criteria for all studies. Most importantly, each set of isomers were fully resolved: delta-8 THCCOOH (*RT*=5.69min); delta-9 THCCOOH (*RT*=5.74min), delta-9 THC (*RT*=8.21min); and delta-8 THC (*RT*=8.65min).

For efficiency, routine DUID cases continue to reflex to the pre-existing 2D LC/MS/MS cannabinoids confirmatory analysis with a 6min runtime. However, if the presence of delta-8 isomers is indicated, the blood specimen is re-analyzed to separate the cannabinoid isomers using the new method. With this new strategy, the cancellation rate decreased from 3.8% to 0.6%, providing forensically defensible results in more than 100 cases.

Delta-8 THC, Isomers, Vaping

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