Cannabis Characterization Using GC and GCxGC with Time-of-Flight Mass Spectrometry

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Introduction

Cannabis is a complex plant containing a variety of naturally occurring organic compounds in a wide range of concentrations, as well as xenobiotics (i.e., polyaromatic hydrocarbons, pesticides, etc.). Among these constituents, the cannabinoids, terpenes, and terpenoids are commonly recognized for their therapeutic properties. This plant has two main subspecies—Cannabis indica and Cannabis sativa—that can be differentiated by their different physical characteristics. Indica-dominant strains are short plants with broad, dark green leaves and have higher cannabidiol content than the sativa plants in which THC content is higher. Sativa-dominant strains are usually taller and have thin leaves with a pale green colour.

The legalization of this drug has also led to an increasing amount of consumer products being infused with CBD, marketed and sold for medicinal and recreational purposes. Thus, it is imperative that we are able to determine its total composition as this is important in determining potency and medical effectiveness (Entourage Effect). In this study, we use GC with Time-of-Flight Mass Spectrometry (TOF-MS) to assist in the determination of its risks and benefits. This powerful tool provides chromatographic separation with high-quality deconvoluted mass spectral data allowing for comprehensive analysis, identification, and quantification of targeted and non-targeted analytes.

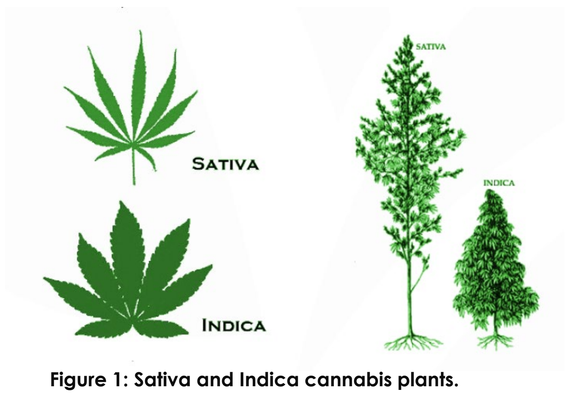


Figure 1: Sativa and Indica cannabis plants.

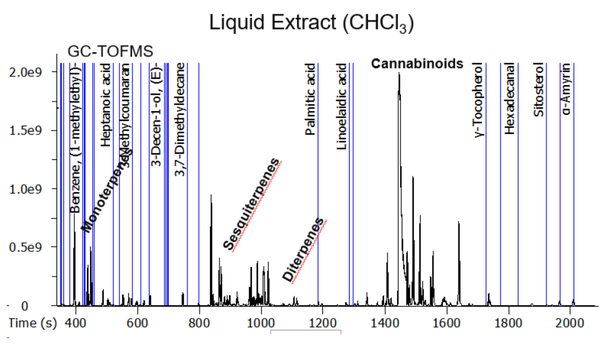


Figure 2: A complex liquid extract of cannabis.

Analytical Challenge

Complexity of Cannabis

1) Dynamic range of compounds.
2) Diversity of cannabis constituents.

Table 1: A complex cannabis sample of a diverse range of constitutes

Name	*Formula*
Ethylbenzene	C_8H_{10}
1-Hexanol	$C_6H_{14}O$
o-Xylene	C_8H_{10}
Heptane, 4,4-dimethyl-	C_9H_{20}
Pentanoic acid	$C_5H_{10}O_2$
2-Heptanone	$C_7H_{14}O$
p-Xylene	C_8H_{10}
Heptanal	$C_7H_{14}O$
5-Hexen-3-ol	$C_6H_{12}O$
Benzene, (1-methylethyl)-	C_9H_{12}
1-Heptanol	$C_7H_{16}O$
Hexanoic acid	$C_6H_{12}O_2$
Benzene, 1,2,3-trimethyl-	C_9H_{12}
Octanal	$C_8H_{16}O$
Heptanoic acid	$C_7H_{14}O_2$
Hexane, 2,3,3-trimethyl-	C_9H_{20}
3-Methylcoumaran	$C_9H_{10}O$

Name	*Formula*
Octanoic acid	$C_8H_{16}O_2$
2-Decanone	$C_{10}H_{20}O$
3-Decen-1-ol, (E)-	$C_{10}H_{20}O$
Dec-(5E)-en-1-ol	$C_{10}H_{20}O$
Nonanoic acid	$C_9H_{18}O_2$
2-Decenal, (E)-	$C_{10}H_{18}O$
3,7-Dimethyldecane	$C_{12}H_{26}$
Hexanoic acid, hexyl ester	$C_{17}H_{34}O_2$
Palmitic acid	$C_{16}H_{32}O_2$
Linoelaidic acid	$C_{18}H_{32}O_2$
Stearic acid	$C_{18}H_{36}O_2$
γ-Tocopherol	$C_{28}H_{48}O_2$
Vitamin E	$C_{29}H_{50}O_2$
Hexadecanal	$C_{16}H_{32}O$
Sitosterol	$C_{29}H_{50}O$
β-Amyrin 3-acetate	$C_{32}H_{52}O_2$
α-Amyrin	$C_{30}H_{50}O$

Alkanes

Aromatics

Alcohols

Aldehydes & Ketones

Acids

Sterols & Tocopherols

Analytical Solution: High Performance TOFMS Instrumentation

- ✓ **Greatly improved sensitivity and linear dynamic range**
- ✓ **Enhanced chromatographic resolution of GCxGC-TOFMS**

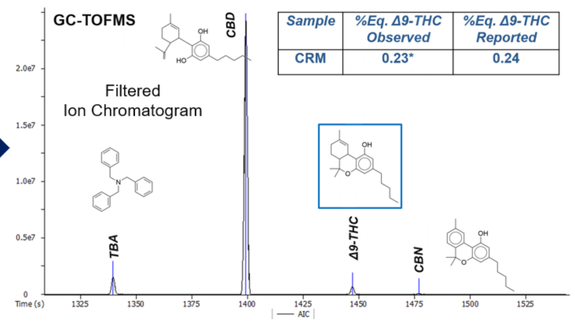
LECO Pegasus® BT 4D

- 1D and 2D Data
- Low fg sensitivity
- 10^5 LDR

Sample Preparation

Purge (N_2), Extract
Anton Paar Monowave
(30 min.)

Collect &
Process Data



Sample	*%Eq. Δ9-THC Observed*	*%Eq. Δ9-THC Reported*
CRM	0.23*	0.24

Results

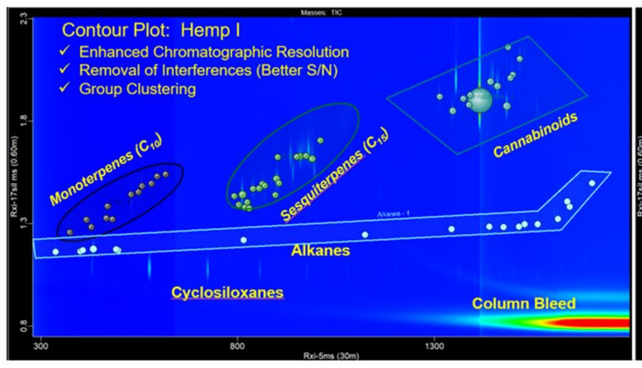


Figure 3: GCxGC-TOFMS contour plot using classifications with m/z filtering.

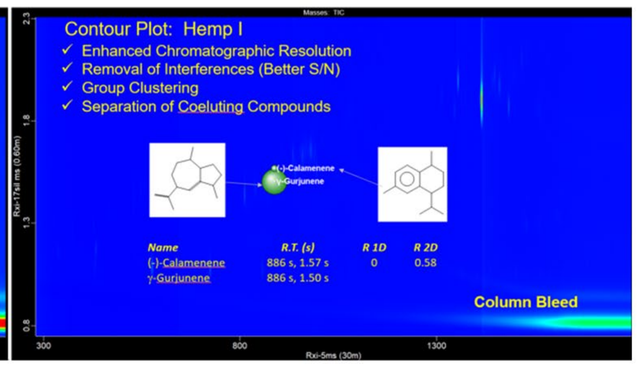


Figure 4: GCxGC-TOFMS contour plot separating coelutions.

Representative Compounds in Hemp

Name	*Formula*	*R.T. (s)*	*Similarity*
α-Pinene	$C_{10}H_{16}$	373.007, 1.259	909
β-Myrcene	$C_{10}H_{16}$	427.012, 1.284	893
Decane	$C_{10}H_{22}$	433.012, 1.177	935
D-Limonene	$C_{10}H_{16}$	466.015, 1.327	871
Eucalyptol	$C_{10}H_{18}O$	469.015, 1.386	833
3-Carene	$C_{10}H_{16}$	481.016, 1.321	813
Linalool, formate	$C_{11}H_{18}O_2$	529.02, 1.387	807
Fenchol	$C_{10}H_{18}O$	547.021, 1.455	916
Isoborneol	$C_{10}H_{18}O$	595.025, 1.531	854
α-Terpineol	$C_{10}H_{18}O$	616.027, 1.542	877
Caryophyllene	$C_{15}H_{24}$	811.042, 1.442	936
α-Bergamotene	$C_{15}H_{24}$	817.043, 1.379	899
α-Guaiene	$C_{15}H_{24}$	823.043, 1.405	823
(E)-β-Famesene	$C_{15}H_{24}$	829.044, 1.375	842
Humulene	$C_{15}H_{24}$	838.045, 1.471	928
Alloaromadendrene	$C_{15}H_{24}$	844.045, 1.470	896
γ-Muurolene	$C_{15}H_{24}$	853.046, 1.472	801
α-Selinene	$C_{15}H_{24}$	868.047, 1.487	889
γ-Gurjunene	$C_{15}H_{24}$	886.048, 1.503	860
(-)-Calamenene	$C_{15}H_{22}$	886.048, 1.569	815

Name	*Formula*	*R.T. (s)*	*Similarity*
(E)-α-Bisabolene	$C_{15}H_{24}$	895.049, 1.441	874
Selina-3,7(11)-diene	$C_{15}H_{24}$	898.049, 1.520	879
α-Calacorene	$C_{15}H_{20}$	901.05, 1.625	805
trans-Nerolidol	$C_{15}H_{26}O$	907.05, 1.457	905
Caryophyllene oxide	$C_{15}H_{24}O$	934.052, 1.629	901
Guaiol	$C_{15}H_{26}O$	940.053, 1.551	870
β-Guaiene	$C_{15}H_{24}$	958.054, 1.629	887
7-epi-a-Eudesmol	$C_{15}H_{26}O$	985.056, 1.654	825
α-Bisabolol	$C_{15}H_{26}O$	994.057, 1.561	889
Olivetol	$C_{11}H_{16}O_2$	1036.06, 1.928	804
Pentadecanal	$C_{15}H_{30}O$	1195.07, 1.416	868
Phytol	$C_{20}H_{40}O$	1246.08, 1.408	874
Cannabidivarol	$C_{19}H_{26}O_2$	1312.08, 1.919	813
Cannabichromene	$C_{21}H_{30}O_2$	1387.09, 1.879	803
Cannabidiol	$C_{21}H_{30}O_2$	1414.09, 1.901	826
Δ9-THC	$C_{21}H_{30}O_2$	1459.09, 1.971	872
Cannabigerol	$C_{21}H_{32}O_2$	1483.1, 1.875	830
Octacosane	$C_{28}H_{58}$	1612.11, 1.324	894

Ave. = 863/1000

- ✓ Terpenes
- ✓ **Monoterpenes (C10)**
- ✓ **Sesquiterpenes (C15)**
- ✓ C20
- ✓ Cannabinoids
- ✓ Alkanes
- ✓ Alkenes
- ✓ Aldehyde
- ✓ Alcohols
- ✓ More…

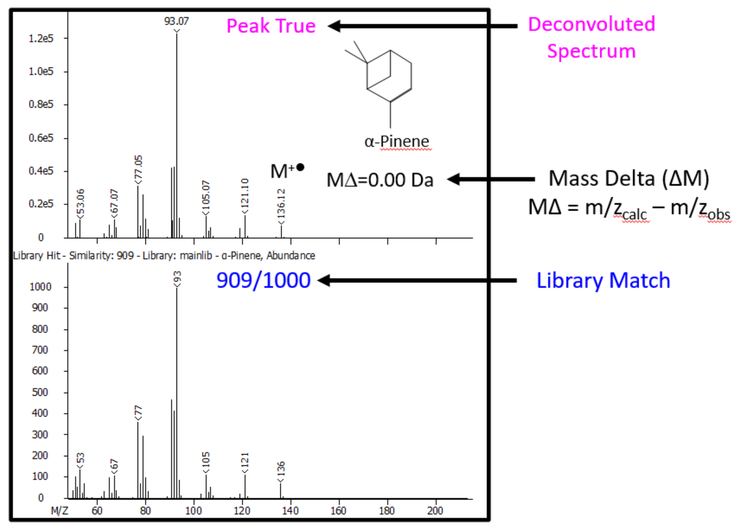


Figure 5: Peak True and Library Match of a-Pinene.

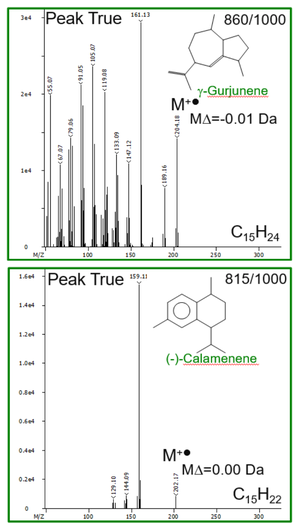


Figure 6: Peak True of separated coeluting compounds.

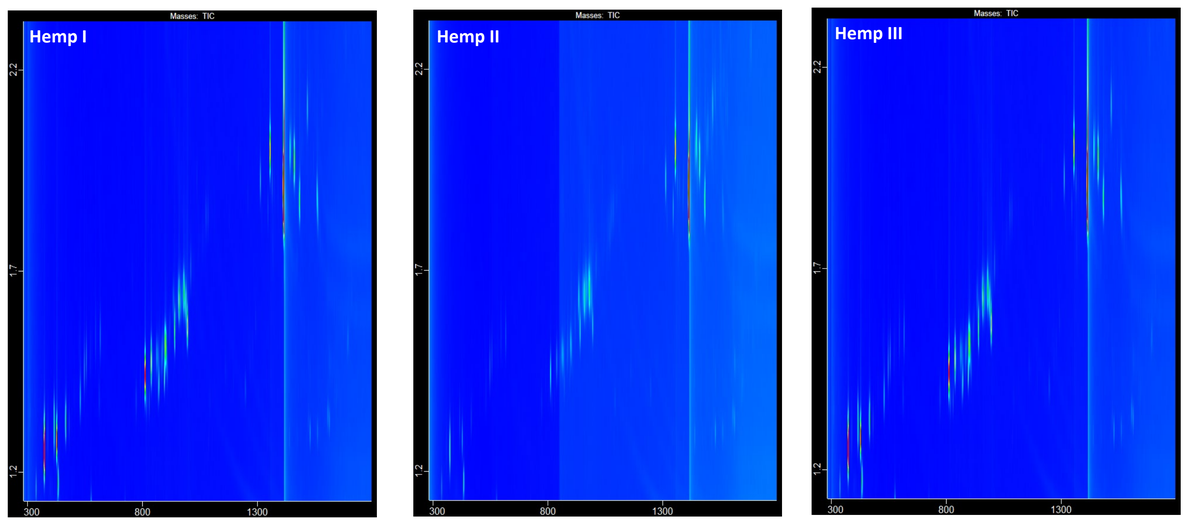


Figure 7: Visual inspection of three hemp maps.

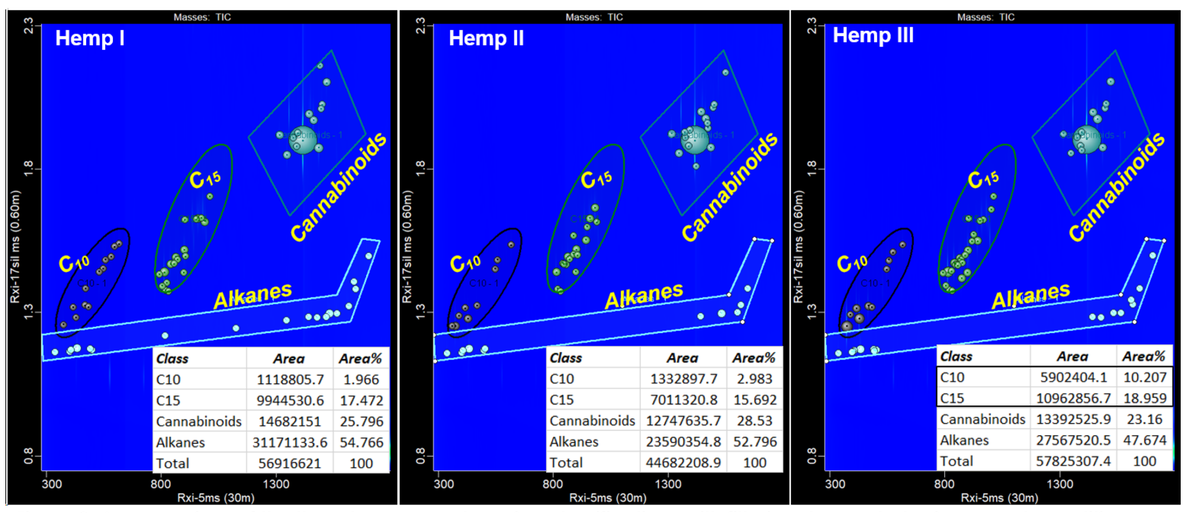
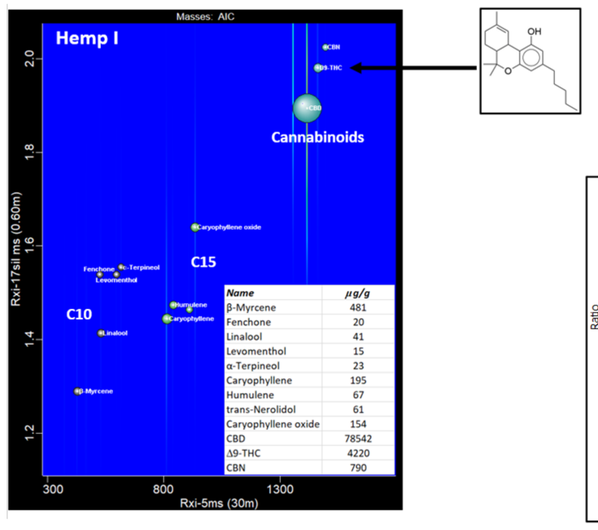


Figure 8: A comparison of the three hemp samples using classifications with filtering demonstrates increased quantities of C10 and C15 terpenes in sample I.



Sample	*%Eq. Δ9-THC Observed*	*%Eq. Δ9-THC Reported*
I	0.42	<0.3

Legal Product? No

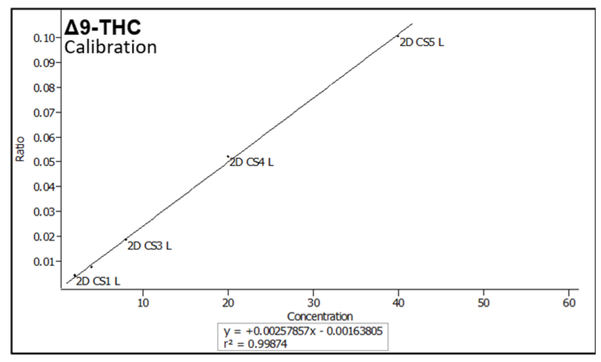


Figure 9: Quantitative analysis of cannabinoids reveals more THC in sample than reported.

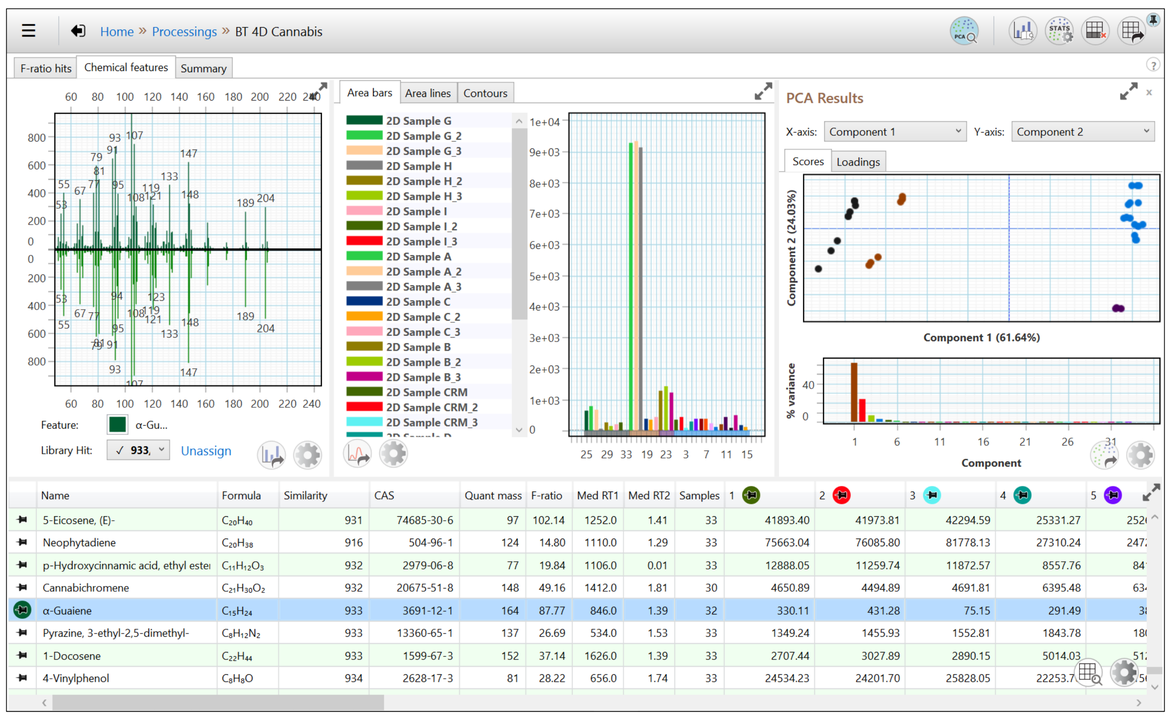


Figure 10: Sample comparison using ChromaTOF® Tile.

Conclusion

GCxGC-TOFMS facilitates compound identification, Library searches, and Mass Δ calculations. Data was used to develop maps for quantitative analysis, cannabis classification, and sample comparison. Cannabis comparison via statistical processing was performed using *ChromaTOF* Tile (based on Fisher ratios) and could also perform PCA analysis.