

Chromatography

Cannabinoid Potency in Cannabis Oil and Medical Marijuana by GC-FID

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Medical marijuana and cannabis derived products are becoming more popular because of legalisation in multiple countries. The increase of this market also increases the offer of products that contain very different levels of cannabinoids than the content described on the label. With more legislation available, the quality control of such products during manufacturing is critical.

Street marijuana usually contains high levels of Δ^9 -THC and a lower level of CBD, whereas medical marijuana and hemp contain high levels of CBD, compared to Δ^9 -THC. The primary psychoactive component is Δ^9 -THC, whilst CBD is the primary therapeutic component.

Consumer hemp generally comes in the form of hemp oil which is used for medical purposes while marijuana is often smoked. All cannabinoid derived products need different sample preparation before analysis due to the difference in sample matrixes.

Potency analysis identifies different cannabinoids and measures their concentration as an indication of the strength of the product. There are over 500 chemical compounds in Cannabis, however there are six target compounds routinely analysed during potency testing. This application note details the identification and quantification of six neutral cannabinoids, by SCION Instruments Gas Chromatography with Flame Ionisation Detection (GC-FID), shown in *Figure 1*. The target compounds were CBD, CBC, Δ^8 -THC, CBG and CBN. For detection of the acidic cannabinoids, primarily THC-A, CBDA, CBDV and CBGA extra sample preparation is required. During sample introduction into a hot injector, decarboxylation of the acidic carboxyl group occurs. To prevent this, and achieve identification of the acidic cannabinoids, derivatisation with BSTFA is recommended.



Figure 1. SCION 4X6 Gas Chromatographs.

Experimental

A SCION 435 GC-FID was used throughout this analysis. However, this application can also be implemented on the 456GC. The instrument can also be equipped with the SCION Instruments Single Quad Mass Spectrometer. An 8400 autosampler was also equipped. The split/split injector was operated at 275°C in split mode. 0.5µL of each analytical standard and sample were injected and separated using the SCION-5MS column. The oven program was 150°C (1 min hold), 15°C/min to 200°C (1 min hold), 15°C/min to 300°C (7

mins hold). The carrier gas was helium at 2mL/min. The FID was operated at a temperature of 300°C. Instrument control and full data processing/reporting was completed on Compass Chromatography Data System.

Analytical reference standards plus a blank were run at a concentration range from 2µg/mL to 100µg/mL. A quality control sample of varying concentrations was also analysed. The concentration of the quality control sample was as follows: CBD 40µg/mL, CBC 30µg/mL, Δ^8 -THC 20µg/mL, 40µg/mL, CBG 15µg/mL and CBN 30µg/mL.

Two different cannabinoid based products were analysed; cannabis oil and medical marijuana. Different sample preparation was required for both samples. The cannabis oil was diluted with methanol before being injected into the GC. The medical marijuana required more sample preparation; the sample was dried for two hours before being ground into small, homogeneous pieces. 30mL of methanol was added and the sample mixture was placed in an ultrasonic bath for 30 minutes. The sample was then evaporated to dryness before being reconstituted in methanol, ready for injection.

Results

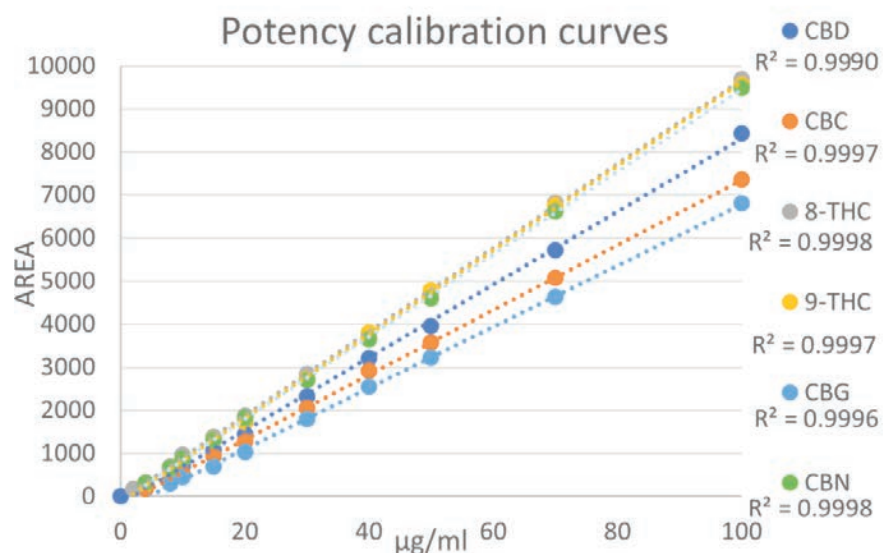


Figure 2. Calibration curve of target compounds.

The calibration curves for the neutral cannabinoids were over a range of 2µg/mL to 100µg/mL. *Figure 2* shows the calibration curve for all target compounds, with excellent linearity observed. All calibration curves exhibited an R² value of >0.999.

The precision of the method was obtained injecting 12 consecutive injections of each target compound at 30µg/mL. The relative standard deviation of each neutral cannabinoid were: CBD 0.50%, CBC 0.39%, Δ^8 -THC 0.38%, Δ^9 -THC 0.44%, CBG 1.89% and CBN 0.24%; highlighting the excellent precision of the method.

Figure 3 shows the chromatogram of a cannabinoid potency reference standard.

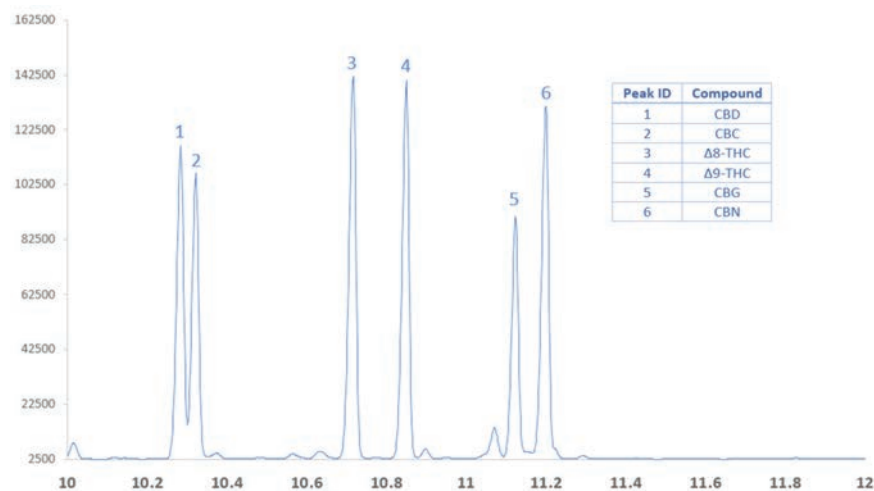


Figure 3. Chromatogram of cannabinoid potency standard.

Figure 3 shows that CBD and CBC are not baseline separated. However, the resolution of the two cannabinoids is 1.2, which enables successful identification and quantification. The peak tailing factor (PTF) of all components was also calculated. The acceptable criteria for PTF is <2. The PTF for all compounds is as follows: CBD 0.83, CBC 0.68, Δ8-THC 0.63, Δ9-THC 0.62, CBG 0.83 and CBN 0.72, highlighting that all compounds met this criteria. In addition, the limit of detection (LOD) and limit of quantitation (LOQ) was also calculated. The LOD's and LOQ's were as follows: CBD 1.5/4.5μg/mL, CBC 0.9/2.75μg/mL, Δ8-THC 0.64/1.95μg/mL, Δ9-THC 0.85/2.65μg/mL, CBG 1.4/4.25μg/mL and CBN 0.73/2.25μg/mL, respectively.

The QC sample was analysed over 5 consecutive injections. The target, actual and RSD% values for the QC sample can be found in Table 1. When analysing cannabinoids or cannabis derived products, it is always good practice to use a blank/ wash in between injections to prevent carryover.

Table 1. Target, actual and RSD% values of QC sample

Compound	Target Value (μg/mL)	Actual Value (μg/mL)	RSD %
CBD	40	37.9	2.04
CBC	30	29.2	1.67
Δ8-THC	20	19.7	0.67
Δ9-THC	40	38	0.64
CBG	15	14.2	2.70
CBN	30	30.1	0.83

The label of the cannabis oil detailed that the sample contains 100mg of CBD (no other ingredients were listed). Analysis of the oil sample confirmed that the actual value of the CBD was 74.2mg with 8.4mg of Δ9-THC. CBC and Δ8-THC was also detected but were

below the LOQ. The medical marijuana did not come with any cannabinoid content listed. The analysis of the sample profiled the cannabinoid content as: 12.5% CBD, 1.25% CBC and 0.7% Δ9-THC. Δ8-THC was detected but was under the LOQ. CBG and CBN were not detected in either sample. Figure 4 shows the chromatograms obtained from the cannabis oil and medical marijuana samples.

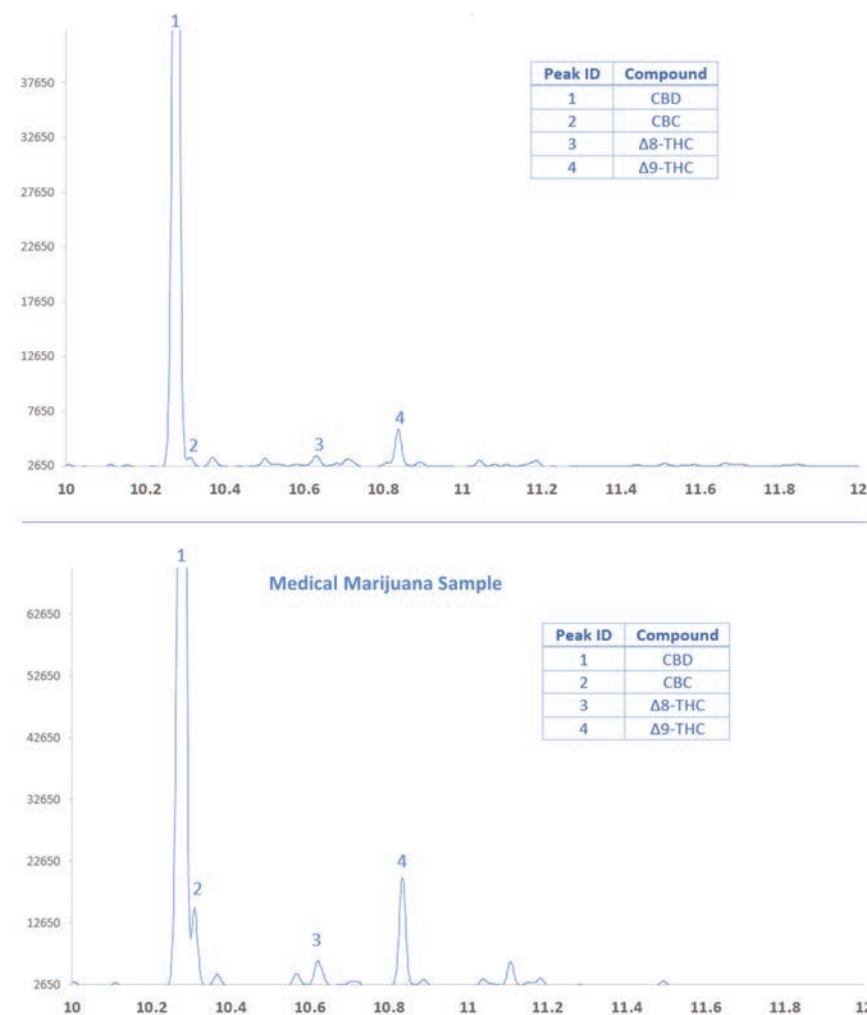


Figure 4. Chromatogram of cannabinoid based samples.

Conclusion

The SCION 4X6 -GC equipped with a split/splitless injector and FID is the ideal solution for the identification and quantification of cannabinoid potency in a variety of samples. This application note demonstrates a simple analysis for the neutral cannabinoids with excellent precision, repeatability and linearity. If both neutral and acidic cannabinoids are to be identified, extra sample preparation is request. Alternatively, the SCION Instruments LC6000 can also be used for potency testing.