APPLICATION NOTE

AN091

Cannabis Potency analysis by GC-FID with helium carrier gas

Scion – Advanced Projects & Custom Solutions

Introduction

Hemp and marijuana are becoming more popular because of legalization 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. Therefore, as quality control it is important to monitor the level of cannabinoids.

Marijuana usually contains high levels of Δ9-THC and a lower level of CBD, with hemp it is the other way around. The primary psychoactive component is Δ9-THC, while 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. Keep in mind that both samples 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, the most tested cannabinoids can be found in table 1. These are also discussed in this application note.

Component	Comment
CBC (Cannabichromene)	
CBD (Cannabidiol)	
Δ8-THC (Δ8-tetrahydrocannabinol)	
Δ9-THC (Δ9-tetrahydrocannabinol)	
CBG (Cannabigerol)	
CBN (Cannabinol)	
CBDV (Cannabidivarin)	Not detectable by GC unless the sample is derivatised.
CBGA (Cannabigerolic acid)	Not detectable by GC unless the sample is derivatised.
THC-A (Δ9-tetrahydrocannabiolic acid)	Not detectable by GC unless the sample is derivatised.
CBDA (Cannabiolic acid)	Not detectable by GC unless the sample is derivatised.

Table 1. Potency components

Injector	Splitless, 275 °C
Column	Scion-5MS
Oven Program	150°C (1 min), 15°C/min to 200°C (1 min), 15°C/min to 300°C (7 min)
Carrier	Helium, 2 ml/min
Detector	FID with ceramic jet, 300°C Air: 300 ml/min, Fuel gas (H_2): 30 ml/min, Make up (N_2): 28 ml/min
Inj. Volume	0.5 µl
Autosampler	8400
Software	Compass CDS

Table 2. Analytical conditions

Experimental

This analysis can be implemented on both the 436-GC as well as on the 456-GC platform. The Scion 436-GC analyser used for this application is equipped with an 8400 autosampler.

The analysis of these components can be done using a GC with a FID detector, but it is also possible to use a Mass Spectrometer (MS). A different method is using a HPLC, in this case it is also possible to detect THC-A, CBDA, CBDV and CBGA with extra sample preparation (derivatization).

When looking at the settings of the GC it is important that the injector temperature is high enough to prevent carry over. Carry over can occur from previous runs or extra material that is being introduced into the system. The chromatogram in figure 1 shows carry over from the previous run. When the injector temperature was increased it showed that the carry over disappeared (see figure 2). Another solution for this problem could be a washing method between samples.

The focus of the calibration lies on six components: CBC, CBD, Δ8-THC, Δ9-THC, CBG and CBN. Unfortunately CBDV, CBGA, THC-A and CBDA cannot be detected without extra sample preparation due to decarboxylation caused by the heat of the injector. Decarboxylation is the removal of a carboxyl group releasing carbon dioxide. CBDV decarboxylates to CBD, CBGA to CBG, THC-A to THC and CBDA to CBD.

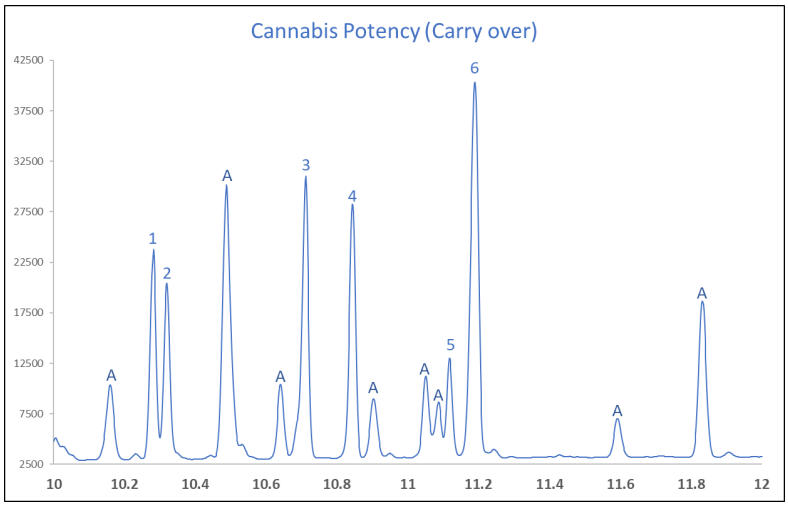

Figure 1. Chromatogram of a potency standard showing carry over (A)

Peak ID	Compound Name
1	CBD
2	CBC
3	8-THC
4	9-THC
5	CBG
6	CBN
A	Carry over peak

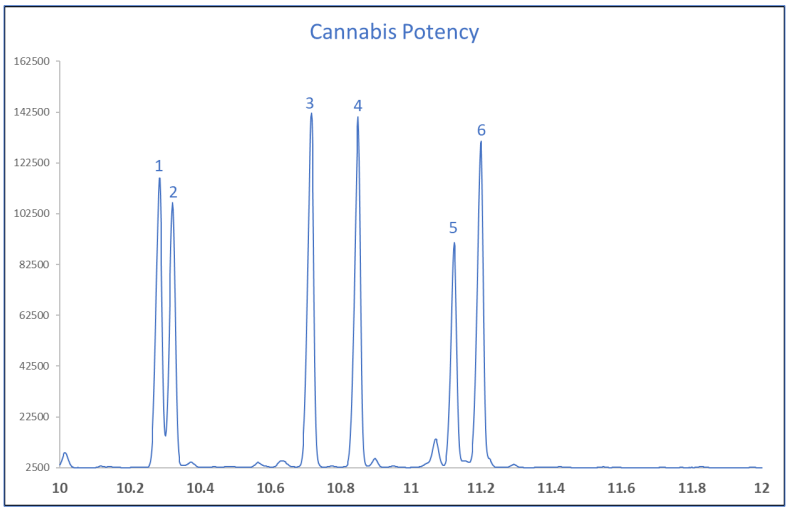

Figure 2. Chromatogram of a potency standard

Peak ID	Compound Name
1	CBD
2	CBC
3	8-THC
4	9-THC
5	CBG
6	CBN

The Quality Control (QC) samples were made from different concentrations of the components: CBD 40 µg/ml, CBC 30 µg/ml, 8-THC 20 µg/ml, 9-THC 40 µg/ml, CBG 15 µg/ml and CBN 30 µg/ml.

Different products were examined to determine potency. Cannabis oil and medical marijuana were chosen in this study.

The cannabis oil was diluted with methanol, for the medical marijuana a bit more was preparation was needed. In order to extract the potency from the medical marijuana, it had to be dried for 2 hours, after that it had to be grinded. 30 ml of methanol was added and placed in an ultrasonic bath for 30 minutes. Evaporate to dryness, reconstitute in methanol.

Results

The precision of the method was obtained by twelve consecutive injections containing 30 µg/ml of each component. Most standards described that the RSD% of an accurate method must be no more than 4%. CBD showed an RSD of 0,5%, CBC 0.39%, Δ8-THC 0.37%, Δ9-THC 0.44%, CBG 1.89% and CBN 0.42%. The precision performance of this method is excellent

When looking at the chromatogram of figure 2 it is shown that CBD and CBC are not base line separated. The two components have a resolution of 1.2, this however is sufficient for accurate quantitation.

In addition to these results, it is shown that the peak tailing factor (PTF) of all components is smaller than 2. CBD 0.83, CBC 0.68, 8-THC 0.63, 9-THC 0.62, CBG 0.83 and CBN 0.72. This overall is an outstanding result.

The calibration curves for the potency standards were prepared between the 2 to 100 µg/ml. Figure 3 shows the calibration range for all the standards and it showed that all the correlation coefficients (R^2) is at least 0.999 which is exceptionally good.

Originally the linearity the limit of detection (LOD) and limit of quantitation (LOQ) was calculated, these results can be found in table 3.

Component	LOD (µg/ml)	LOQ (µg/ml)
CBD	1.5	4.5
CBC	0.9	2.7
8-THC	0.64	1.9
9-THC	0.85	2.6
CBG	1.4	4.2
CBN	0.73	2.2

Table 3. LOD and LOQ for the different components

The QC sample was analysed in four runs of each five injections. The average concentration for CBD was 37.9 with an RSD% of 2.04, CBC was 29.2 with a RSD% of 1.67. 8-THC was 19.7 with an RSD% of 0.67, 9-TH was 38 with an RSD% of 0.64, CBG was 14.2 with an RSD% 2.70 and CBN was 30.1 with an RSD% of 0.83.

The analysis of two cannabis samples was performed on an oil and a medical marijuana sample.

According to the label of the oil sample it should contain 100 mg of CBD, other components were not mentioned on the label. After analysis it showed that the oil sample contained 74.2 mg CBD and 8.4 mg 9-THC. CBC and 8-THC were also detected, unfortunately these were below the LOQ, it also shows that CBG and CBN were not present in this sample.

The medical marijuana did not come with an index stating it's content. Analyses showed it contained 12.5% CBD, 1.25% CBC, 0.7% 9-THC. 8-THC was detected but the concentration was below the LOQ, CBG and CBN were not present in this sample.

Figure 4 shows an example chromatogram of the oil and medical marijuana sample.

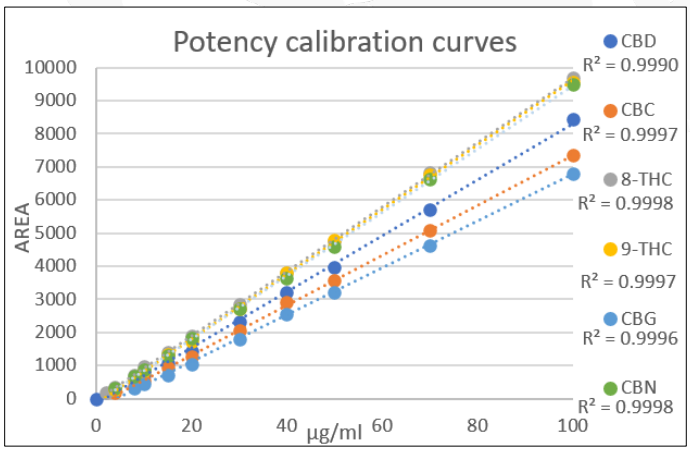

Figure 3. Calibration curves of the potency standards

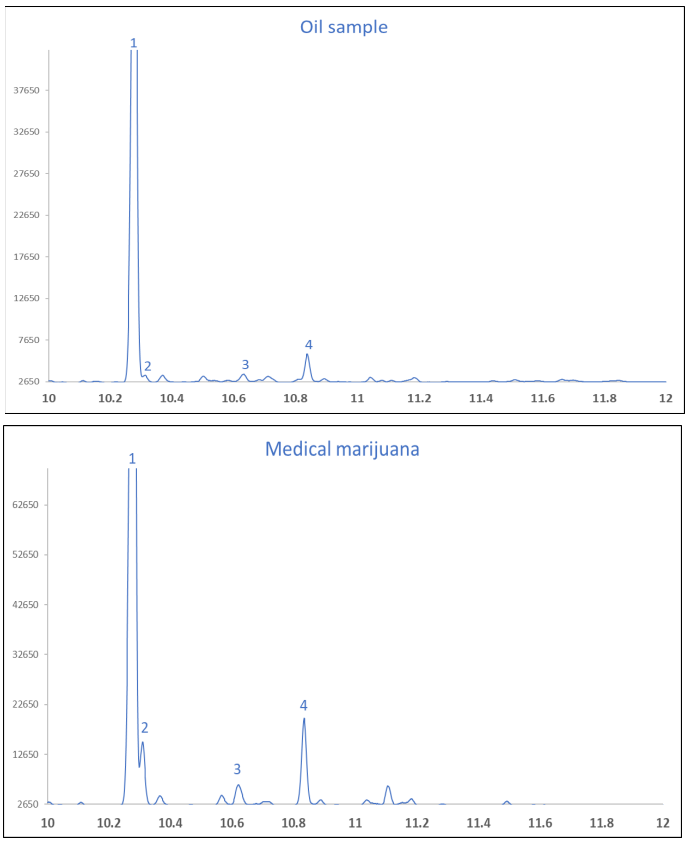

Figure 4. Chromatogram of an oil and medical marijuana sample

Peak ID	Compound Name
1	CBD
2	CBC
3	8-THC
4	9-THC
5	CBG
6	CBN

Conclusion

The Scion 4X6-GC analyser equipped with an split/spitless injector, scion instruments column and FID is capable of analysing potency from cannabis products in a qualitative and quantitative way.

Although the method developed is well suited for potency analysis it is confirmed that one of the disadvantages of this method is that the acidic form of the potency components is converted in the hot injector. It should be kept in mind that the levels of CBD, CBG and 9-THC in this case is a summation of the non-acidic and acidic form. If the acidic form has to be analysed it is advised to perform extra sample preparation or another analysing method in the form of a Scion 6000 HPLC method can be chosen.

Picture 1: The Scion 4X6-GC series with 8400 autosampler

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