1. Introduction

Both hemp and marijuana are varieties of the cannabis plant. While marijuana generally possesses high levels of the psychoactive compound tetrahydrocannabinol (THC), hemp is just the opposite: rich in CBD but low in ∆9-THC. While more research is needed to better understand the chemistry of benefits from CBD, it has been reported to reduce convulsions, inflammation, nausea and anxiety, and has even eradicated tumors in some patients. (Note: the cannabinoid(s) recommended for specific medical conditions have not been approved by the FDA).

CBD-rich oil has become increasingly popular and is administered via sublingual drops, gel capsules or as a topical ointment. The main source of CBD-rich oil is industrial hemp. CBD oil is derived as concentrate from CO2 or butane extraction of hemp, sometimes followed by steam distillation or ethanol distillation for purification.

The FDA has issued warning letters to firms that market unapproved new drugs allegedly containing CBD. As part of these actions, the FDA has determined the cannabinoid content of some hemp products and many were found to contain levels of CBD that are very different from the label claim. It is important to note that such products are not approved by the FDA for the diagnosis, cure, mitigation, treatment, or prevention of any disease.

Therefore, as quality control, the level of CBD and THC in any such products should be monitored. Like cannabis, hemp oil may be analyzed easily and effectively for its cannabinoid content. In this report, both THC and CBD levels in two different hemp oil samples were determined using a GC-FID setup.

2. Experimental Methods

The cannabinoids standard of CBD, ∆9-THC and CBN (cannabinol) was purchased from Restek (cat no. 34014). Hemp oil samples were obtained from various vendors.

To make calibration standards, original stock of a cannabinoids standard was diluted in methanol to indicated concentrations. The hemp oil samples were diluted as follows: 10uL of oil was first diluted into 400uL isopropanol, mixed thoroughly, then diluted again into 400uL methanol and mixed thoroughly.

Analytical Conditions

<table>
<thead>
<tr>
<th>Column</th>
<th>SH-Rxi-5Sil MS, 15m x 0.25mm x 0.25µm (part no. 227-36036-01)</th>
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</thead>
<tbody>
<tr>
<td>GC oven temp</td>
<td>200°C ramp at 15°C/min to 300°C, hold 5min</td>
</tr>
<tr>
<td>SPL</td>
<td>250°C; Hydrogen carrier gas, constant inlet pressure at 48.3kPa, split ratio = 10</td>
</tr>
<tr>
<td>FID</td>
<td>300°C; Hydrogen flow rate 32mL/min; Air flow rate 200mL/min; Nitrogen makeup gas flow rate 24mL/min.</td>
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</tbody>
</table>

Data were acquired and analyzed using Lab Solutions software.
3. Results and Discussion

A six-point calibration curve was generated using the cannabinoid standards of 0.8ppm, 4ppm, 20ppm, 100ppm, 250ppm and 500ppm each of CBD, ∆9-THC and CBN in methanol and fitted to linear regression (fig.1). The correlation coefficient ($r^2$) values of all three compounds are >0.999 over 6 standard levels.

1. CBD (cannabidiol)

2. THC (Δ9-tetrahydrocannabinol)

3. CBN (cannabinol)

<table>
<thead>
<tr>
<th></th>
<th>CBD (total)</th>
<th>THC (total)</th>
</tr>
</thead>
<tbody>
<tr>
<td>conc (%)</td>
<td>%RSD</td>
<td>conc (%)</td>
</tr>
<tr>
<td>Sample 1</td>
<td>2.182</td>
<td>0.893</td>
</tr>
<tr>
<td>Sample 2</td>
<td>0.062</td>
<td>0.555</td>
</tr>
</tbody>
</table>

Table 1: Measured concentrations of CBD and THC in two different hemp oil samples.

Each oil sample was diluted as described in Experimental Methods and run as triplicates. CBN was not detected in either of these samples. To calculate the % level of cannabinoids in the samples, the reported ppm concentration of the sample from LabSolutions software was multiplied by 81 (dilution factor) then divided by 10,000 (10,000ppm = 1%).

Figure 1: Standard curves for cannabinoids

Figure 2: Example chromatogram of hemp oil sample 1. Note that CBN was not detected in the sample.
4. Conclusions

In this report, cannabinoid CBD and THC levels in two hemp oil samples were analyzed by the Shimadzu GC-2030 with FID and AOC-20i autoinjector. Calibration was carried out from 0.8ppm to 500ppm for each cannabinoid with excellent linearity. No carryover was observed with this setup and method. H₂ was used as the carrier gas for efficient and low cost analysis. Furthermore, with an integrated hydrogen sensor option for GC-2030, the instrument can be used with hydrogen carrier gas routinely with minimal safety concerns for the operator.

As shown in the table of results above, sample 1 is shown to contain a high level of CBD (2.18%), while sample 2 has very little CBD in comparison (0.06%). Clearly not all products are created equal. And this signifies the importance of assaying CBD level in these claimed CBD-rich products. It should be noted, though, that the CBD amount reported in the sample is the summation of CBD and CBDA (Cannabidiolic acid), or total CBD, as the CBDA present would have been converted to CBD in the hot injection port of the GC and added to the CBD peak.

Also shown by the results above, both hemp oil samples contain very low levels of the psychoactive compound THC. The low level of THC (< 0.3%) is what distinguishes hemp from marijuana; therefore, the THC level is another important parameter to monitor in these products. Similar to CBD, one consideration is that the acid form of THC known as THCA will be converted to THC in the hot injection port of the GC; as a result, the THC level reported above is the sum of THCA and ∆9-THC, or total THC.

Furthermore, this method can be adapted easily to assay the THC level in medical marijuana for potency. However, as mentioned above, the level determined will be the sum of THCA and ∆9-THC. If individual compound concentration is desired, an HPLC method should be employed instead. Information regarding Shimadzu HPLC cannabis analyzer can be found on the Shimadzu website ssi.shimadzu.com.