

Medical Cannabis Potency Testing using the SRI 8610C FID GC

The SRI 8610C FID GC with a 12-vial sample heater is designed for testing the **potency of medical cannabis (cannabinoids)**. With minor configuration and procedural changes the GC can also test for **terpenes** and **residual solvents** in concentrates (see our documents on our website at www.srigc.com/documents.htm).

The 12-vial sample heater aids in a quicker extraction of the cannabinoids in solvent and maintains the extracted samples at 50° C for better reproducibility.

The GC includes SRI's Flame Ionization Detector (FID) which is able to measure the cannabinoid molecules based on its ability to detect the combustion of hydrocarbon molecules.



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The cannabinoid molecules, **Δ 9-THC**, **CBD**, and **CBN** (and for more advanced operators, **CBC**, **Δ 8-THC**, and **CBG**) are separated by a 15-meter metal capillary column which is heated in the column oven.



Hook up the gas lines to the left side of the GC. The GC can be operated with hydrogen or helium as a carrier gas. When using hydrogen as a carrier gas, cap off the hydrogen gas inlet and connect the hydrogen to the carrier 1 inlet.

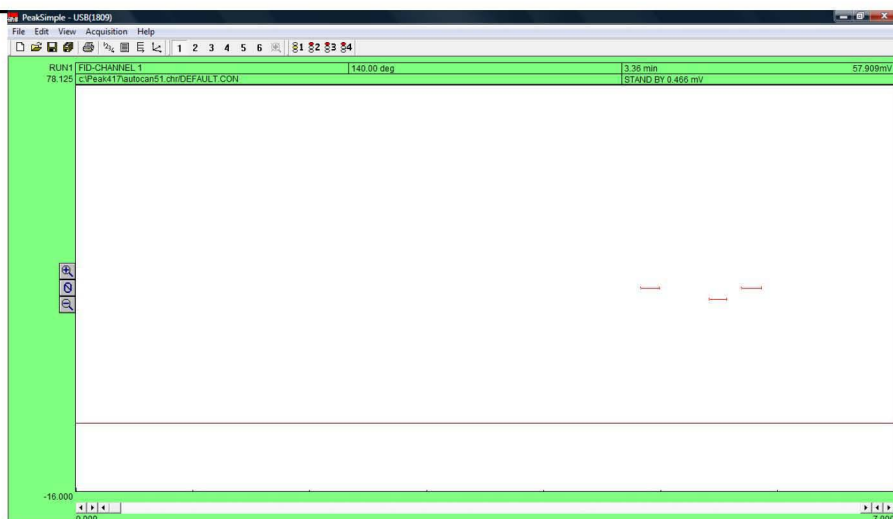


The entire GC plugs into any Windows computer using a USB cable.

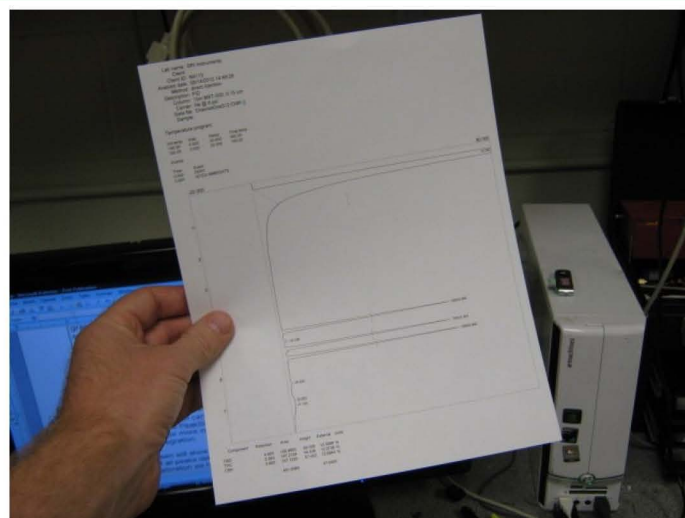


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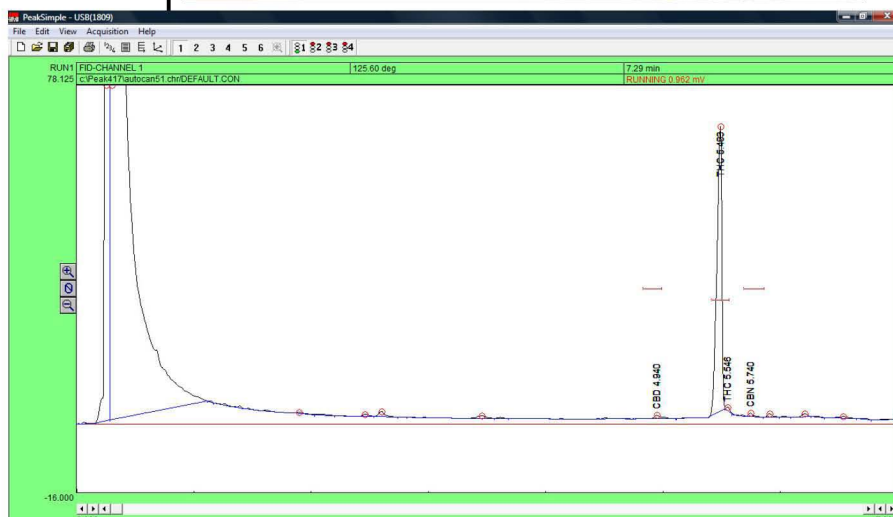
SRI's PeakSimple software is included with the GC. PeakSimple software collects the GC data and generates a calibrated result which can be printed or transferred to other programs such as Excel or Word.



The chromatogram hardcopy print-out at right shows the three peaks CBD, THC and CBN which were injected to calibrate the GC.



An actual cannabis sample is shown at right.

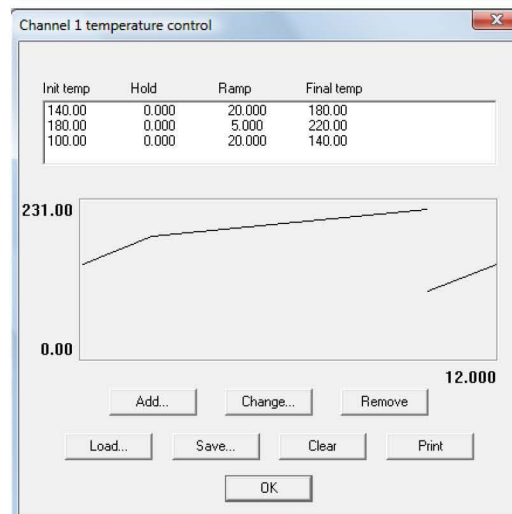
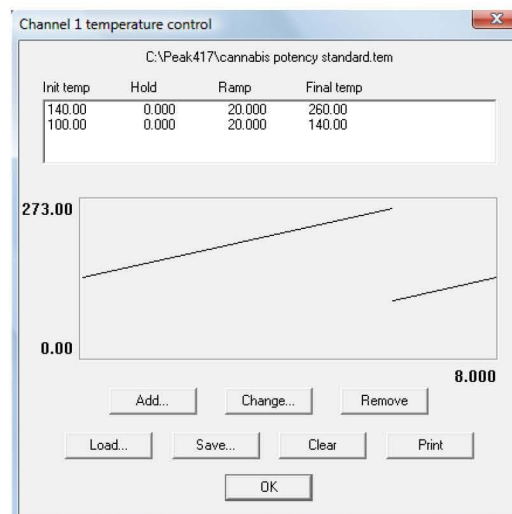


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For a quick 8-minute analysis that optimizes speed *and* peak separation, set the column oven temperature as shown to the right.

Sometimes, a better separation is preferred (particularly with THC, CBG, and CBN) at the expense of speed. For a longer 12-minute analysis, set the column oven temperature as shown to the right.

Set the integration parameters as shown. Note the "Sample weight" box. When you calibrate the GC it will be set at 100. When you run actual cannabis samples, the weight of the sample will be entered. (ex. If the sample weighed 0.104 grams, then "104" should be entered).



Channel 1 integration

Peak detection sensitivity

Peak: 95.00 %

Base line: 60.00 %

Area reject: 1

Standard weight: 4.000

Spike channel

☒ None

☐ 1

☐ 2

☐ 3

☐ 4

☐ 5

☐ 6

Merge results from channels:

☐ 1

☐ 2

☐ 3

☐ 4

☐ 5

☐ 6

Sample weight: 100.000

OK Cancel

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Obtain the cannabinoid calibration standard from a chromatography supplier like Restek (restek.com). The standards can be acquired individually, but SRI recommends a more convenient three-way (THC, CBD, CBN) cannabinoid standard. The standards are available at a concentration of 1000 ng/ul in Methanol. No license is required to purchase.

Break the glass ampoule and transfer the contents into a 2mL septum vial. Restek provides one free vial with each standard.

Whether you have three vials (individual standards of THC, CBD, or CBN) or one vial of 3-way standard, they will each be at a concentration of 1000ng/ul. We will refer to these standards as primary standards. Ideally, when not in use they should be kept in a refrigerator with an unpierced septum so that the methanol will not evaporate and increase the concentration of the cannabinoids in the standard. When calibrating with the primary standards the percent concentration of the cannabinoids will be approximately 40%.



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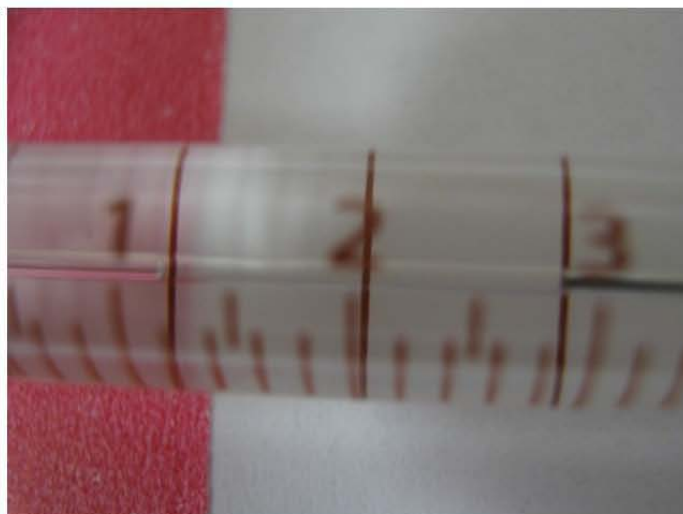
Pull the plunger back to the 3ul mark and note the amount of liquid. It should be 1.6-1.8ul because the needle also contains .6-.8ul and this adds to the 1ul you measured with the plunger.

Leave the plunger at the 3ul mark.

With the plunger still at the 3ul mark, place the needle up against the septum (but not poking through it yet).

Press the Start Run button or hit the Spacebar on the keyboard to start the run.

Insert the syringe all the way through the septum as far as it will go. Immediately depress the plunger. Twist the syringe one half turn (to wipe off any liquid on the tip of the needle) and then withdraw the syringe.



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SRI recommends preparing a "333 working standard" rather than using a primary standard to calibrate. Not only will this help to preserve the purity of your primary standard and get more mileage out of it, but it will also calibrate the GC at percent concentrations that more closely resemble cannabis flowers (13.32% instead of 40%).

If you have separate cannabinoid standards, use the 100uL syringe, which is included with the SRI GC, (Restek#24863) to transfer 100uL of each 1000ng/uL (primary) standard into another 2mL vial. If you have the 3-way standard, use the 100uL syringe to transfer 100uL of the standard into another 2mL vial and then add 200uL of methanol.

After either method, you will end up with 300uL of working standard containing 333ng/uL each of the three compounds (CBD,THC and CBN). Label the primary and working standards with both a name and a date.

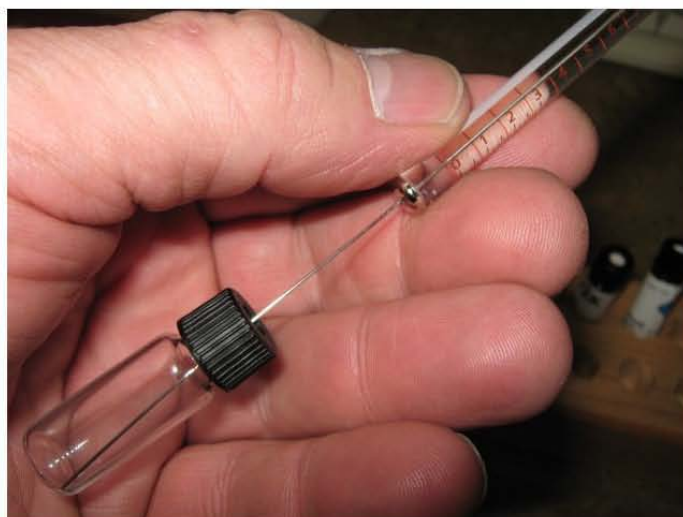


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Rinse the syringe first then use the 10ul syringe delivered with the GC (SRI #8670-9550) to withdraw 2-3ul of the working standard. Puncture the septum rather than open the vial to avoid letting the methanol solvent evaporate each time the vial is opened. Pump the plunger several times to get rid of air bubbles.

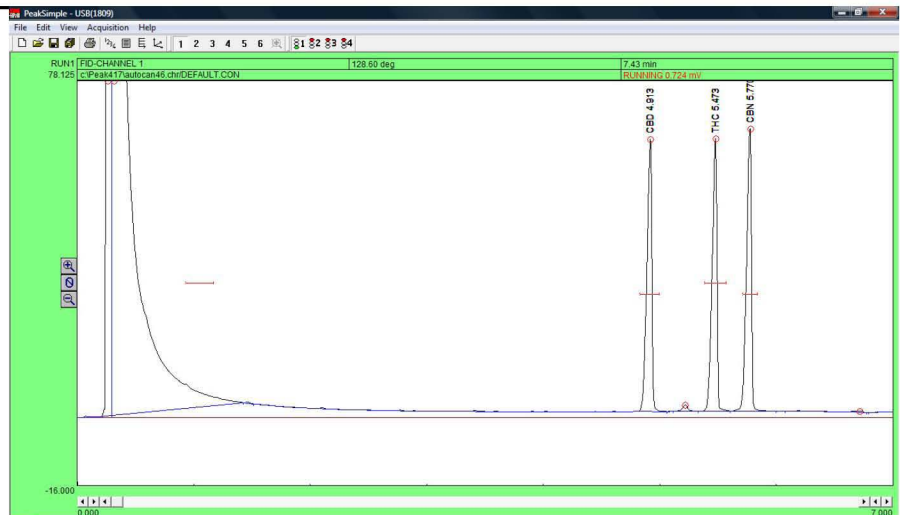
With 2-3ul of liquid in the syringe, hold the needle vertically or at least slanted upwards so any air bubbles will rise towards the needle.

With air bubbles removed, push the plunger to the 1ul mark. It is important to be as precise as possible. Wipe the needle with your fingers or a tissue to remove any liquid from the outside of the needle.



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Once the run is completed you should see a large solvent peak near the beginning, then closer to the end, three peaks of roughly equal size (there will also probably be a small Delta-8 THC peak between the 1st and 2nd peak). Add retention windows to the three peaks by right clicking on the peak and selecting "Add component". See the PeakSimple tutorial describing the process of creating retention windows.



Identify the three peaks (from left to right: CBD, THC, CBN) by right-clicking on each peak and selecting "Edit component". Assign each peak a unique number and name (CBD, THC, or CBN), select "show largest peak only", and add a "%" sign to the "Units" box. Press the "OK" button to exit back to the main chromatogram screen.

The 'Component details' dialog box is shown with yellow arrows pointing to specific fields. The 'Peak number' is set to 2. The 'Peak name' is 'THC'. The 'Start' time is 5.39 and the 'End' time is 5.57. The 'Units' field contains a percentage sign (%). The 'Internal standard' is 0.000. The 'Internal standard peak' is 0. The 'Ref peak' is 0. Under 'In case of multiple peaks', 'Show largest peak only' is selected. Under 'Measure peak', 'Area' is selected. The 'Multiplication factor' is 0.00000000. The 'Calculate area as time-slice' checkbox is unchecked. The 'OK' and 'Cancel' buttons are at the bottom.

Right click on the chromatogram and select "Components" to open the "Channel 1 Components" Screen. Here will be displayed a list of all the components with named retention windows and unique peak numbers. Select "Save" and name the component file so that if you exit PeakSimple your component and calibration files will not be lost.

The 'Channel 1 components' dialog box shows a table of components. The table has columns for Peak, Name, Start, End, and Calibration. The components listed are CBD, THC, and CBN. Below the table are buttons for Add..., Change..., Remove, Calibrate..., Load..., Save..., Clear, and Print. The 'OK' button is at the bottom.

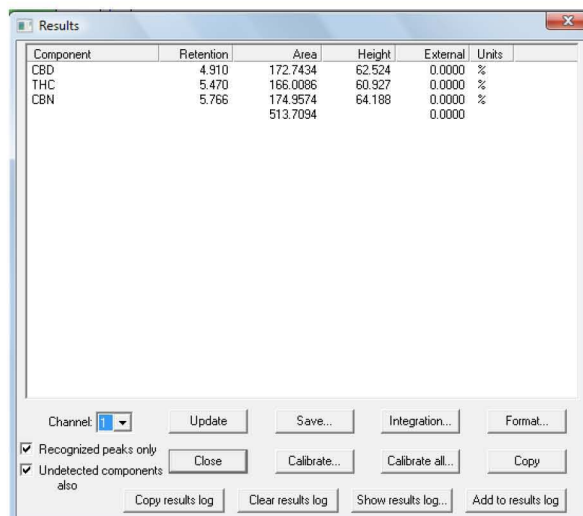
Peak	Name	Start	End	Calibration
1	CBD	4.833	4.999	CBD Cal cal
2	THC	5.389	5.569	THC cal cal
3	CBN	5.715	5.842	CBN cal cal

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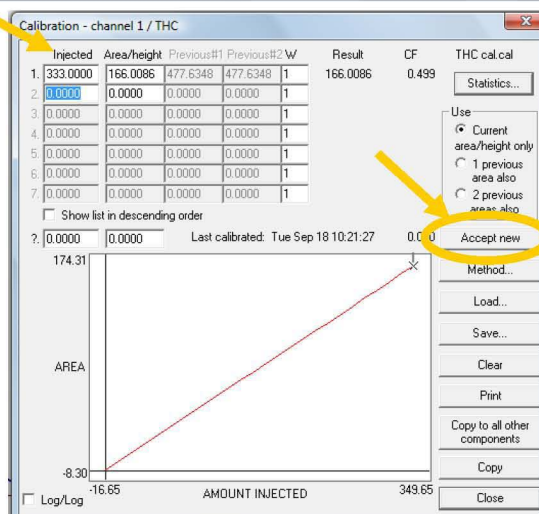
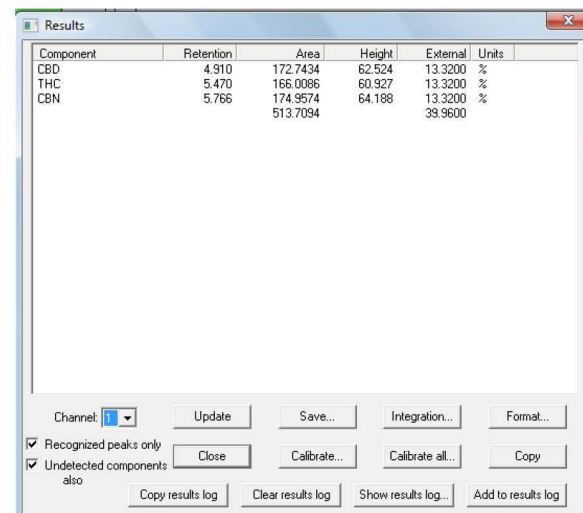
Check the Results screen. If you injected a primary standard the area counts for the cannabinoids should be between 420 and 540 and roughly equal to each other (+/- 30). If you injected the working standard the area counts should be between 140 and 180 and roughly equal (+/- 10).

Calibrate each peak by creating a calibration curve. See the Peak-Simple tutorial describing this process. In the calibration curve enter the amount of standard you just injected. This will be 333 (for 333ng/ul) or 1000 (for 1000ng/ul). Type this number in the top left cell of the spreadsheet in the calibration curve. Then click the Accept New button to transfer the peak's area into the top row 2nd column. Save the curve under some name. Do this for all the peaks.

Navigate to the View/Results screen to see the report. With the integration screen and components setup as discussed earlier in the document the percent concentrations of CBD, THC, and CBN will each be displayed as 13.32% (or 40% if primary standards were injected). You are now calibrated and ready to inject real cannabis samples.



Component	Retention	Area	Height	External	Units
CBD	4.910	172.7434	62.524	0.0000	%
THC	5.470	166.0086	60.927	0.0000	%
CBN	5.766	174.9574	64.188	0.0000	%
		513.7094		0.0000	

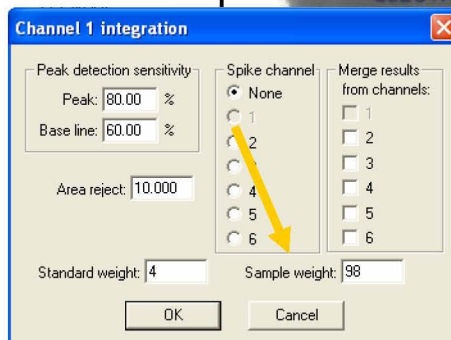
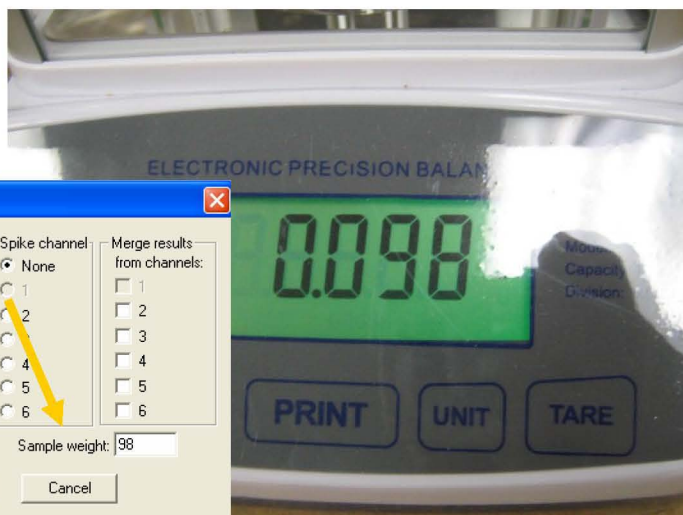
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		513.7094		39.9600	

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Remove the cap from a 40ml vial and place it on the balance. The balance should be capable of reading 1 milligram (.001 gram). A balance like this can be purchased brand new for less than \$300 on E-bay.

With the 40 mL vial on the balance tare the reading (make the reading 0.000). Then carefully add 100 milligrams of manicured cannabis. Drop the bits of cannabis into the vial slowly until the reading is close to 100 milligrams. Make sure to write down the exact weight of the sample somewhere, preferably on the vial itself.

Don't worry if you are slightly under or above 100. In the photo at right, the reading is 98 milligrams which is close enough. You will enter the reading in the sample weight field in Peak-Simple software which will mathematically correct the calculated answer to compensate for weights slightly above or below 100.

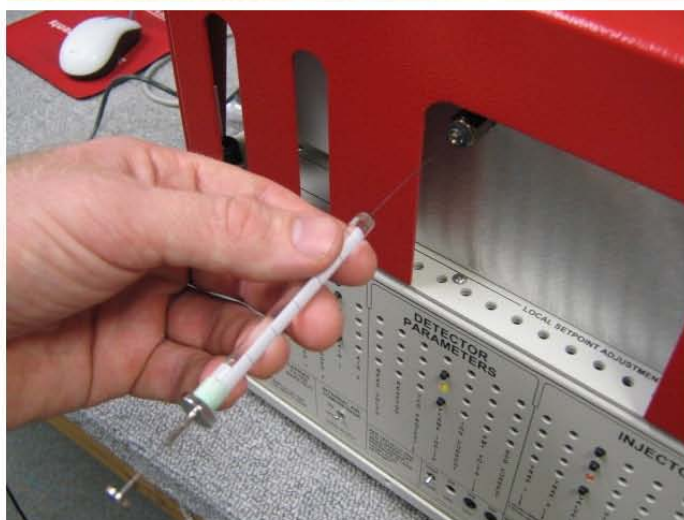
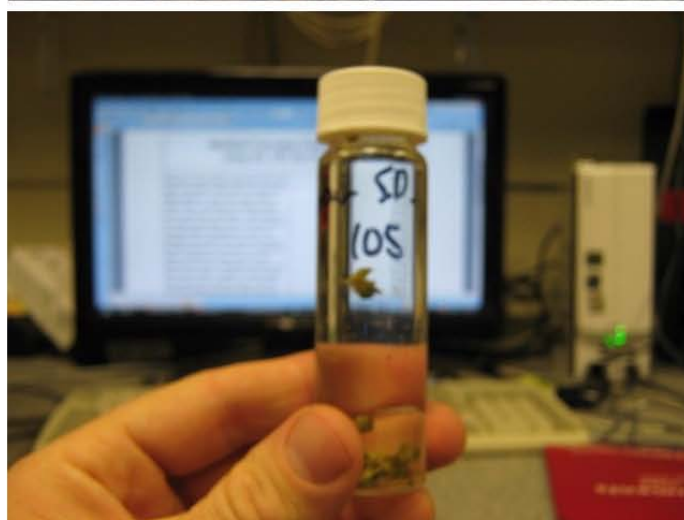


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Remove the 40ml vial from the balance and fill it with 40ml of extraction solvent. You can use 70% or 91% IPA, methanol (methyl alcohol), ethanol, acetone, chloroform or other solvents. We recommend using either methanol, or for a cheap and efficient solvent, denatured alcohol (a mixture of ethanol and methanol) that can be obtained at most hardware stores for less than \$20 a gallon. Non-polar solvents like hexane are not recommended because they do not extract the cannabinoids as well as polar solvents.

Shake the vial for a few seconds and then let it sit for about 20 minutes in the incubator (longer without heat).

Use the 10ul syringe which comes with the GC to inject 1ul of the extract as shown previously with the calibration standard. It is important to be very precise with the syringe since the overall accuracy of the test depends on this. Don't forget to enter the exact Sample weight in the proper field on the integration screen.



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A real cannabis sample will look something like the chromatogram at right. There will be one big peak (THC) and much smaller ones for CBD and CBN. In this case, CBD is so low that it is not detected.

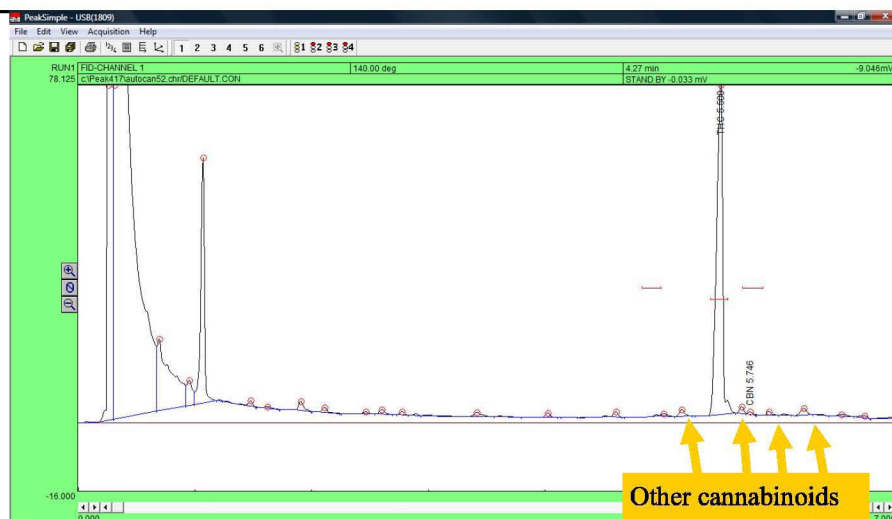
CBN may or may not be detected or it may blend into the much larger THC peak. When this happens you can use the slower temperature program and/or lower the carrier pressure to get better separation of the peaks.

There may be other peaks which are not CBD, THC or CBN. These other peaks are cannabinoids (CBC, Delta-8 THC, CBG, and others) for which there may or may not be calibration standards available at this time.

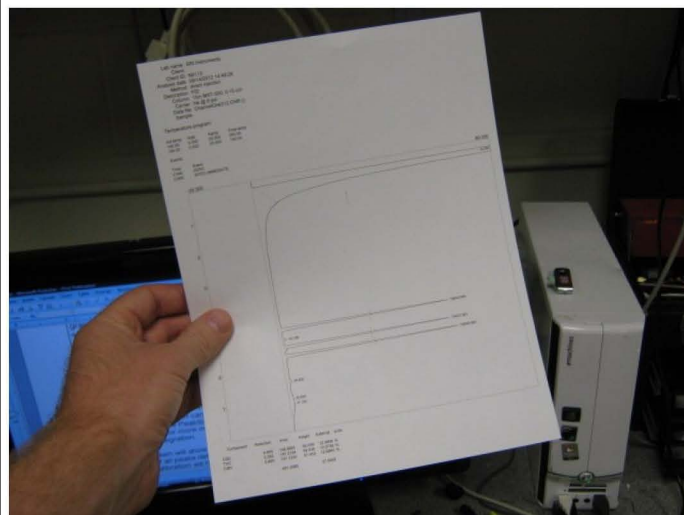
It may be necessary to manually integrate some of the peaks for the most accurate quantification of cannabinoid potency. See the PeakSimple Advanced Tutorial for more information on manual Integration.

The Results screen will show the concentration of all peaks detected based on the calibration we have previously done.

Print the chromatogram and results for a hardcopy record of the analysis.

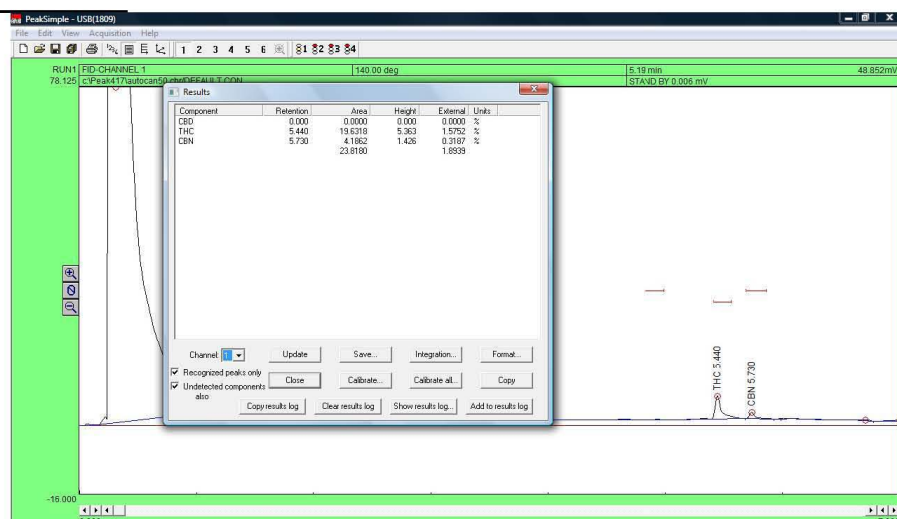


Component	Retention	Area	Height	External	Units
CBD	0.000	0.0000	0.000	0.0000	%
THC	5.500	273.1762	85.229	21.9188	%
CBN	5.746	1.1584	0.496	0.0882	%
		274.3346		22.0070	

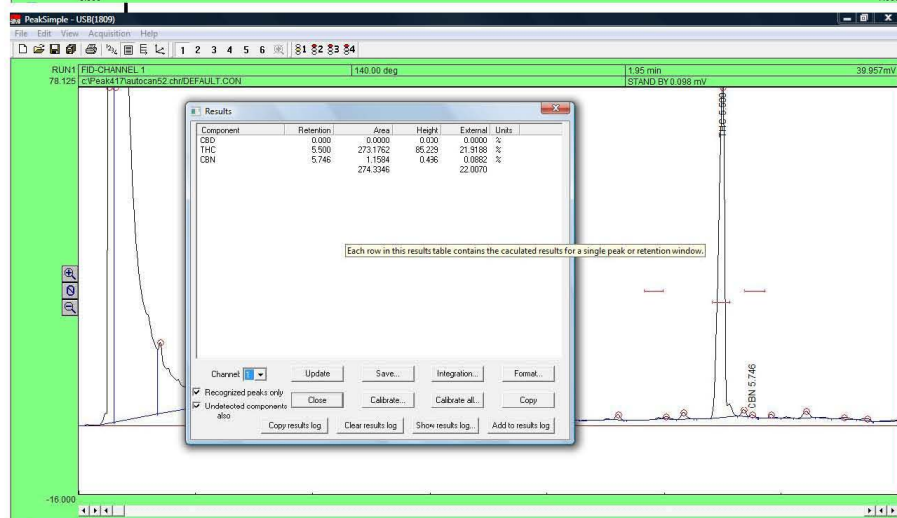


Medical Cannabis Potency Testing using the SRI 8610C FID GC

Chromatogram of a low-potency cannabis flower sample with 1.6 % THC.



Chromatogram of a high-potency cannabis flower sample with 21.9 % THC.



Chromatogram of a typical cannabis concentrate with 40.2% THC.

