



# **TENNESSEE BUREAU OF INVESTIGATION**

## *Forensic Services Division*

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### Forensic Chemistry Standard Operating Procedure Manual Tetrahydrocannabinol Concentration Screening

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#### **35.0 TETRAHYDROCANNABINOL CONCENTRATION SCREENING PROCEDURE FOR PLANT MATERIAL, SUSPECTED CANNABIS LIQUIDS, VAPE CARTRIDGES, AND “GUMMY” EDIBLES USING GC/MS**

##### **35.1 Application**

This procedure is used to compare tetrahydrocannabinol (THC) concentrations in suspected cannabis, vape cartridge, cannabis liquid, and gummy edible exhibits to a 1% concentration THC standard. This procedure does not determine the exact concentration of THC in these suspected exhibits. Full quantitation procedure for plant material only is discussed in Chapter 36.

Gummy edibles will be referred to as gummies for ease of reading throughout this chapter.

##### **35.2 Exhibit Requirements for Analysis**

35.2.1 Any plant material exhibits that have not been previously analyzed by the TBI FCU should be tested according to Chapter 25 prior to screening.

35.2.2 Vape cartridges, suspected cannabis liquids, and gummies that have not been previously analyzed by the TBI FCU should be run on the GC/MS to determine if THC is present prior to conducting a concentration screen. A Duquenois – Levine test should be run in conjunction with the GC/MS.

35.2.3 If the analyst must destroy a vape cartridge in order to obtain the liquid within, they will repackage the broken cartridge in a puncture-proof container and place the liquid in a vial for exhibit preservation.

##### **35.3 Equipment and Reagents**

###### **35.3.1 Equipment**

- Gas Chromatograph Mass Spectrometer
- Analytical Balance
- Disposable glass tubes with screw caps
- Disposable transfer pipettes
- Disposable autosample vials, inserts, and caps
- Vortexer
- Grinding apparatus (plant material only)
- Assorted volumetric glassware
- Hot plate (for gummies)
- Glass beakers (for gummies)
- Volumetric pipettes with disposable tips
- Syringes and syringe filters (for gummies)



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#### 35.3.2 Reagents

- Methanol (for plant material and vape cartridges)
- Hexanes (for gummies)
- High purity water (for gummies)
- Tribenzylamine
- $\Delta^9$ -Tetrahydrocannabinol standard (1mg/mL)

#### **35.4 Plant material testing preparation and procedures**

##### 35.4.1 Internal standard preparation (300 $\mu$ g/mL Tribenzylamine)

Weigh 30 mg of tribenzylamine on the analytical balance and dissolve into 100 mL of Methanol using a volumetric flask. Other equivalent ratios may be used to prepare a 300  $\mu$ g/mL tribenzylamine solution.

##### 35.4.2 Comparison standard preparation (THC)

Pipette 500  $\mu$ L of internal standard and 500  $\mu$ L of THC standard into a disposable tube and vortex until a homogenous solution is obtained.

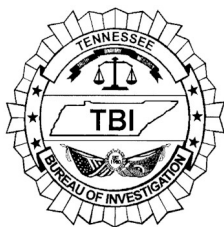
##### 35.4.3 Sample preparation

1. Exhibits should have a beginning net weight of 1.0 gram or more for screening and sample preservation as discussed in section 11.8. If less than 1.0 gram is available, the analyst will consult their supervisor or the customer on how to proceed.
2. After collecting a representative sample for analysis, grind the plant material until a relatively homogenous mixture is formed.
3. Weigh approximately 100 mg of plant material on an analytical balance. Record this weight to two decimal places for percent THC calculations after instrumental analysis.
4. Pipette 1 mL of 300  $\mu$ g/mL internal standard solution and 1 mL of methanol onto the plant material.
5. The sample will be vortexed for at least ten minutes.
6. The plant material must remain in the extraction solution for a minimum total of 30 minutes before analysis.

#### **35.5 Vape cartridge and suspected cannabis liquid testing preparation and procedures**

##### 35.5.1 The procedure for plant material will be used for vape cartridges with the following modifications.

1. Approximately 100 mg of liquid is needed. If the analyst does not have enough exhibit to obtain 100 mg and leave enough for independent testing, they will consult their supervisor or the customer on how to proceed.
2. Tare the glass extraction tube and weigh the liquid sample directly into the tube.
3. If needed, the liquid may be heated in the oven at 80°C or in a warm water bath to help reduce the viscosity for sample collection.



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4. Pipet 1 mL of methanol mixed with 1 mL methanol/tribenzylamine internal standard in to the liquid sample. Vortex the sample until completely dissolved. Extended extraction times are not required since most vape liquid dissolve quickly
5. Samples can be diluted with methanol if necessary. The analyst must account for dilution with any additional volume of methanol when performing the calculations. While this step is optional, it may be necessary to prevent carry-over on the instrument.

#### **35.6 Gummy edible testing preparation and procedures**

35.6.1 The procedure for plant material will be used for gummy edibles with the modifications listed in 35.6.2 through 35.6.4.

35.6.2 Internal standard preparation (300 µg/mL Tribenzylamine)

Add 30 mg tribenzylamine into 100 mL of Hexane. This internal standard solution should be treated in the same manner as the methanol version outlined in 35.4.1.

35.6.3 Comparison standard preparation (THC)

Prepare the 1 % THC standard in hexane using the same methodology as outlined in 35.4.2

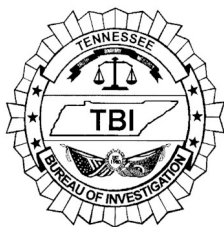
35.6.4 Sample preparation

1. Place approximately a 100 mg piece of gummy into 3 mL of high purity water. Record the weight of the gummy piece prior to dissolving for calculations.
2. If the analyst does not have enough exhibit to obtain 100 mg and leave enough for independent testing, they will consult their supervisor or the customer on how to proceed.
3. Heat the gummy and water mixture in a flat-bottomed glass tube in a water bath near boiling until the gummy piece has completely dissolved into the water.
4. Pipet 1 mL of hexane and 1 mL of the hexane/tribenzylamine internal standard solution into the aqueous gummy mixture once it has cooled.
5. Vortex this extraction mixture for 30 minutes. Extract the organic layer for analysis.
6. Filtering the extract with a syringe filter is highly recommended for this procedure since not all of the protein matrix will denature during heating. Filtering will be required if the hexane mixture appears to have emulsified during the extraction.

#### **35.7 Instrumentation**

35.7.1 The GC/MS data will be collected with the appropriate validated method.

35.7.2 The liner and septum for the GC/MS will be changed immediately before running any screening standards and samples.



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#### **35.8 Quality Assurance**

- 35.8.1 Procedural blanks will be run between all standards and samples. These blanks must be free of tribenzylamine and THC for an acceptable autosample run.
- 35.8.2 Clean or disposable glassware will be used in all sample and standard preparations. Disposable pipette tips will be used when pipetting and discarded after use.
- 35.8.3 The same internal standard preparation must be used for both the THC standard and the sample preparations.
- 35.8.4 The methanol/tribenzylamine internal standard will be verified by GCMS prior to use to ensure no contaminants are present and will have a one (1) year expiration date. Usage will be tracked in same manner as other in-house reagents.
- 35.8.5 The hexane/tribenzylamine internal standard will be freshly prepared prior to analyses.
- 35.8.6 The 1% comparison standard can only be compared to the samples that were pipetted by the same analyst on the same day with the same pipette.
- 35.8.7 The initial threshold used for integration must be recorded in the casefile and must be identical for the comparison standard and the sample.

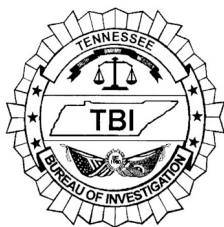
#### **35.9 Performance Verification and Acceptance Criteria**

- 35.9.1 The instrument should produce an adequate response relative to the baseline on the TIC for both tribenzylamine and THC in order to use generated data.
- 35.9.2 Refer to the GCMS Chapter for criteria regarding acceptability of the mass spectrometer for use.
- 35.9.3 The comparison standard ratio must be within the acceptable ratio range located in the 1% THC standard acceptability range document located in the Lab Documentation folder in Ensur.

#### **35.10 Interpretation**

- 35.10.1 THC mass spectra must be evaluated for the correct fragmentation pattern.
- 35.10.2 The ratio of ion abundances (correlated areas) for THC versus tribenzylamine will be calculated by dividing the THC abundance by the tribenzylamine abundance for both the standard and the sample.
- 35.10.3 In order to report greater than 1% for THC, the abundance ratio of the sample must be above the abundance ratio of the standard.
- 35.10.4 The analyst will need to correct the abundance ratio to account for variations in starting sample weight as demonstrated below:

$$\text{Corrected abundance ratio} = \text{abundance ratio} (100 \text{ mg} / \text{sample weight})$$



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#### 35.10.5 Example Calculation:

##### ❖ Example 1:

The analyst obtained the following data from this procedure

	<i>THC standard</i>	<i>Sample</i>
<i>THC abundance</i>	29616555	32465544
<i>Tribenzylamine abundance</i>	10462257	7947738
<i>Abundance ratios</i>	2.831*	4.085

In the example, the analyst weighed out 101.34 mg of plant material. The analyst will then apply the above calculation to correct for this weight.

$$\begin{aligned}\text{Corrected abundance ratio} &= 4.085 (100 \text{ mg} / 101.34 \text{ mg}) \\ &= 4.085 (0.9868) \\ &= 4.031 \text{ (rounded to three decimal places)}\end{aligned}$$

Since the sample's abundance ratio (4.031) is greater than the standard ratio (2.831), then the sample has a THC concentration greater than 1%.

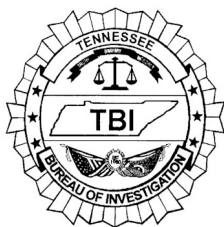
\*For this example, the standard ratio is assumed to be in the acceptable range as outlined in 35.9.3. Consult Ensar for current acceptability ranges.

35.10.6 The analyst will consult with their supervisor or technical leader if the sample's corrected abundance ratio is equal to or less than the comparison standard's abundance ratio.

#### 35.11 Reporting

35.11.1 Plant material exhibits that do not have a prior report released that have greater than 1% total THC will be reported as follows. A separate report for presumptive cannabis testing will be issued in addition to this report.

<u>EXHIBIT(S):</u>			
001-a	Plant material		
<u>RESULTS:</u>			
	<u>Controlled Substance</u>	<u>Total THC Percentage</u>	<u>Amount</u>
001-a	delta-9-tetrahydrocannabinol	Greater than 1%	152.20 grams



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35.11.2 Plant material exhibits that have greater than 1% total THC, that have been previously reported as cannabis, and the entire previously reported amount was screened, will be reported as follows.

<u>EXHIBIT(S):</u>			
001-a	Plant material		
<u>RESULTS:</u>			
	<u>Controlled Substance</u>	<u>Total THC Percentage</u>	<u>Amount</u>
001-a	delta-9-tetrahydrocannabinol	Greater than 1%	-

35.11.3 Plant material exhibits that have greater than 1% total THC, that have been previously reported as cannabis, and only part of the previously reported amount was screened, will be reported as follows. Note that all weights reported are weights recorded from the initial report.

<u>EXHIBIT(S):</u>			
001-a	Plant material		
<u>RESULTS:</u>			
	<u>Controlled Substance</u>	<u>Total THC Percentage</u>	<u>Amount</u>
001-a	delta-9-tetrahydrocannabinol	Greater than 1%	152.20 grams
	Screening was not performed on the remaining plant material previously reported as cannabis.		

35.11.4. Vape cartridges and gummy exhibits that have greater than 1% total THC will be reported as follows. The descriptions of the evidence may vary in order to best describe the exhibit(s).

<u>EXHIBIT(S):</u>			
001-a	Vape cartridge		
001-b	Gummy candy		
<u>RESULTS:</u>			
	<u>Controlled Substance</u>	<u>Total THC Percentage</u>	<u>Amount</u>
001-a	delta-9-tetrahydrocannabinol	Greater than 1%	1 Unit
001-b	delta-9-tetrahydrocannabinol	Greater than 1%	1 Unit



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35.11.5 Other suspected cannabis liquids will be report as follows.

<u>EXHIBIT(S):</u>			
001-a	Liquid (either weight or approximate mL)		
<u>RESULTS:</u>			
	<u>Controlled Substance</u>	<u>Total THC Percentage</u>	<u>Amount</u>
001-a	delta-9-tetrahydrocannabinol	Greater than 1%	-

35.11.6 The above examples will also adhere to the reporting requirements regarding testing requirements and supplemental reports outlined in the Reporting Chapter.