



Chemistry and Testing of Metabolite Hemp

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Specialized Metabolism

Also called:

- Natural Products
- Secondary Metabolite

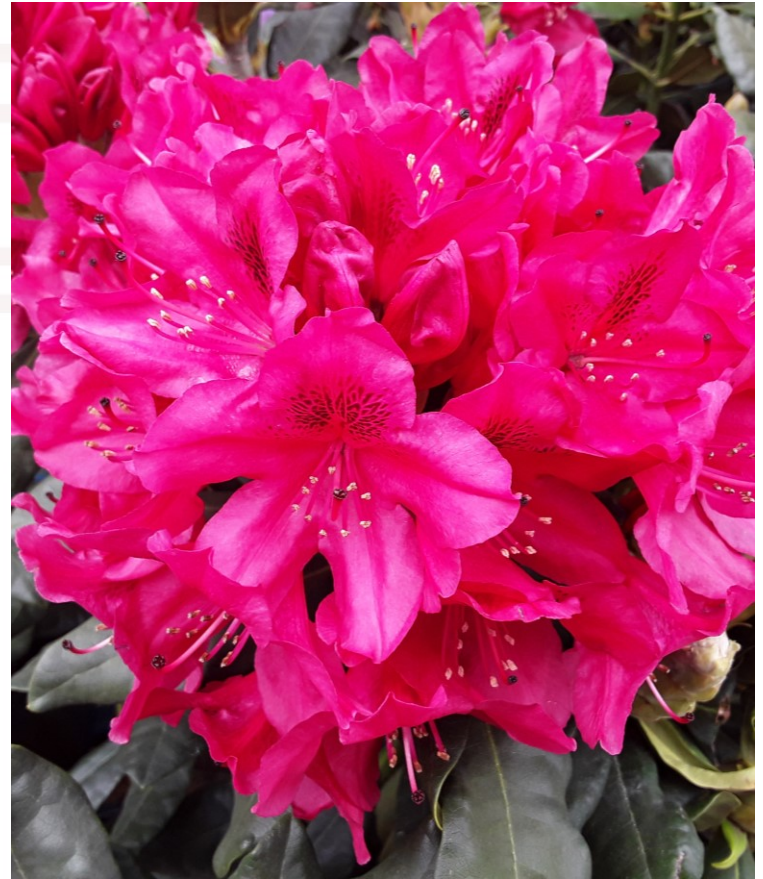


Compounds which are non-essential for an organism's growth or development but helps improves its ability to survive

Specialized Metabolites Roles

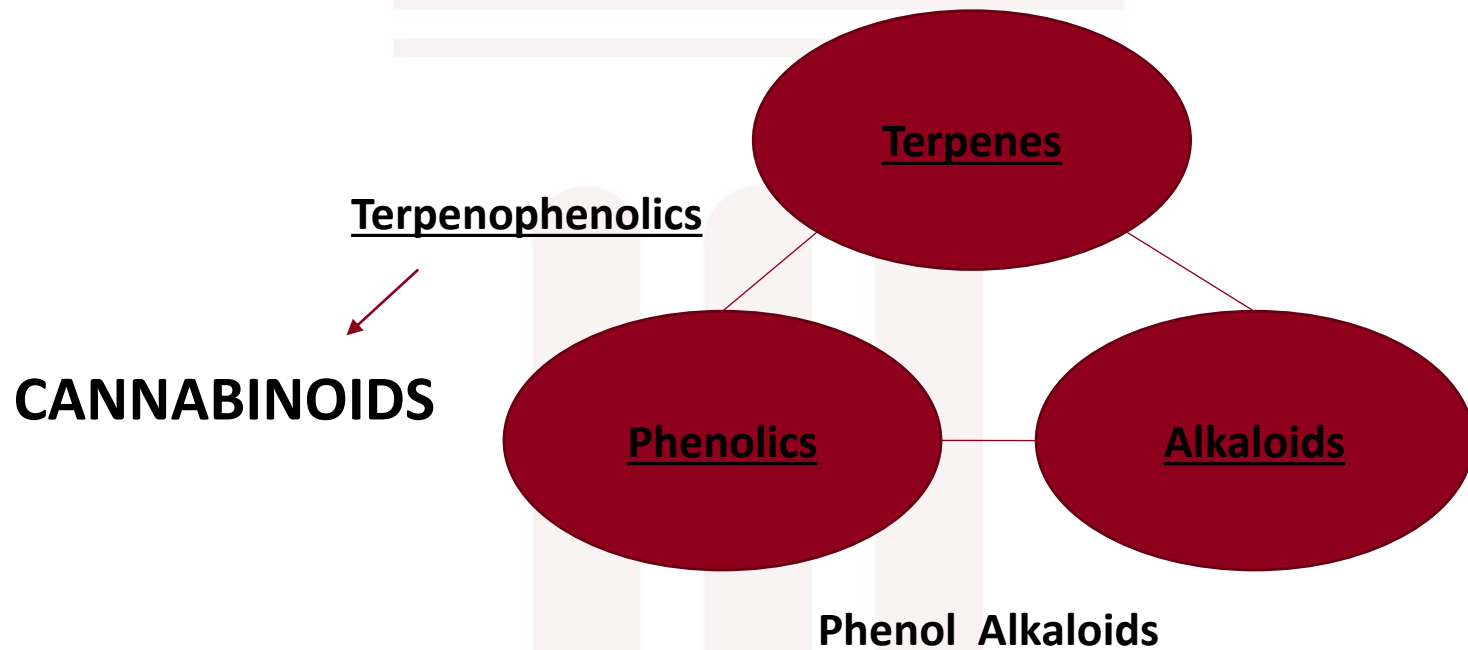
Plants synthesize specialized metabolites to:

- Deter or kill herbivore pests
- Deter or kill pathogens
- Deter nearby plants
- Attract pollinators or seed dispersers
- Attract predators of herbivores
- Protect against environmental stresses



Cannabis Chemistry

- Cannabis is an amazing chemical biosynthetic factory, both in quality and quantity of specialized metabolites



Defining Hemp

- Hemp is *Cannabis sativa* and any part of that plant, including the seeds thereof and all derivatives, extracts, cannabinoids, isomers, acids, salts, and salts of isomers, whether growing or not, with a Δ^9 -tetrahydrocannabinol (THC) concentration of not more than 0.3% on a dry weight basis.

-- Agriculture Improvement
Act of 2018

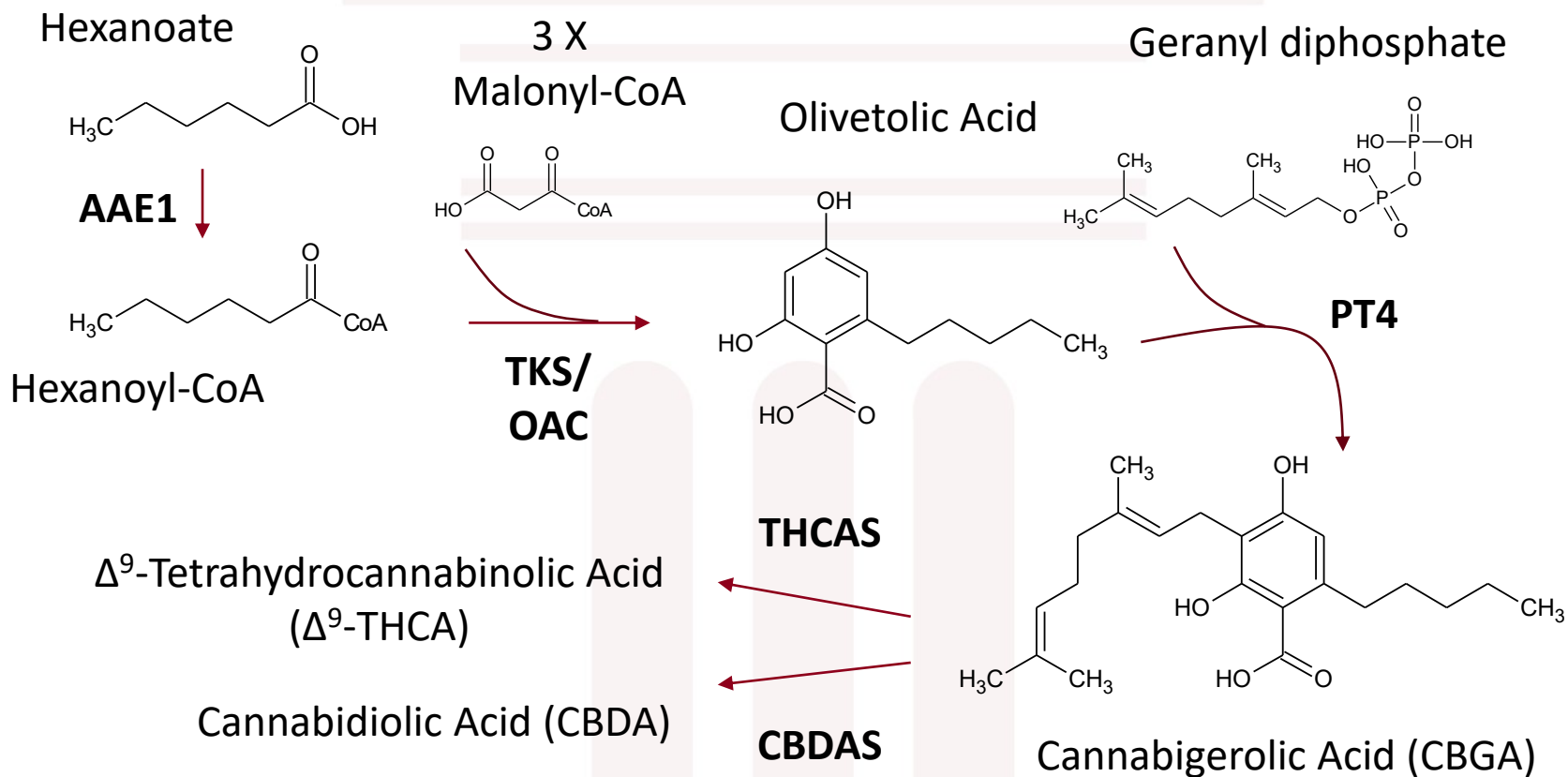


Cannabinoid Production

- Cannabinoids are produced in trichomes
- Primarily on bracts of female flowers

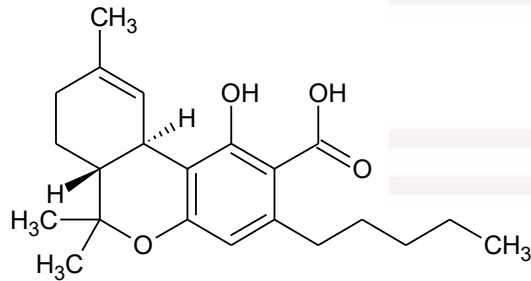


Biosynthesis of Cannabinoids

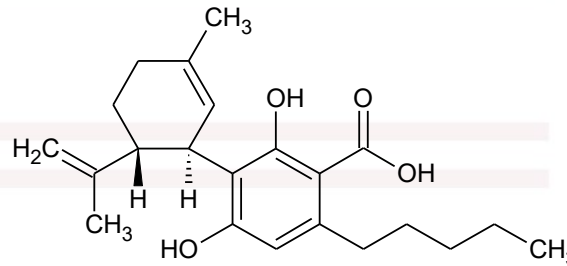


Cannabis Chemistry

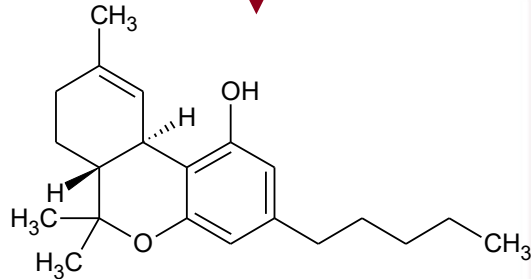
Δ^9 -THCA



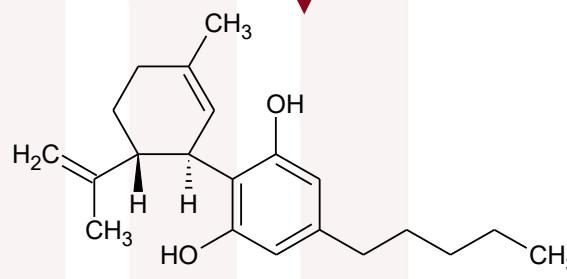
CBDA



Acidic form
(produced by
the plant)



Δ^9 -Tetrahydrocannabinol (Δ^9 -THC)

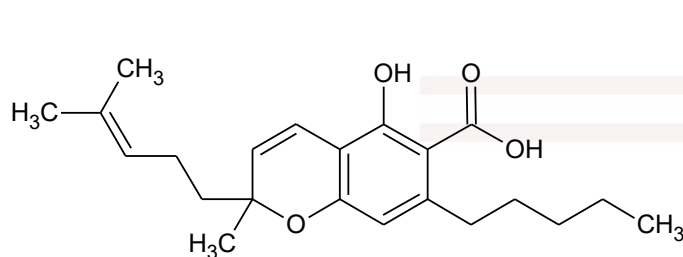


Cannabidiol (CBD)

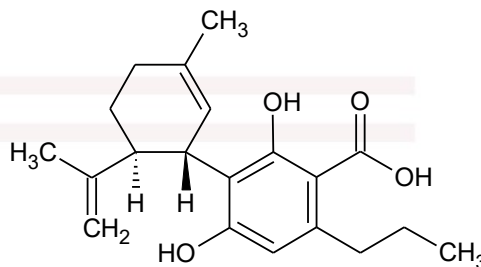
Neutral form
(typically
very little in
the plant)

Other Cannabinoids

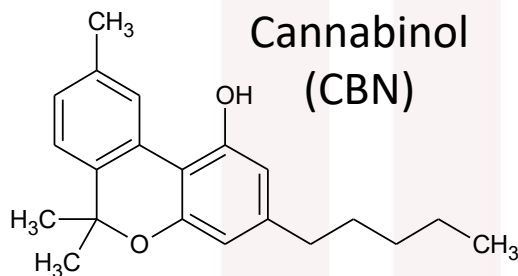
- Cannabis produces >120 cannabinoids



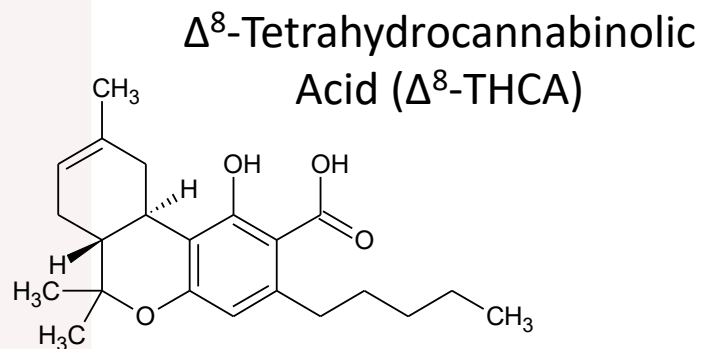
Cannabichromenic Acid
(CBCA)



Cannabidivarinic
Acid (CBDVA)



Cannabinol
(CBN)

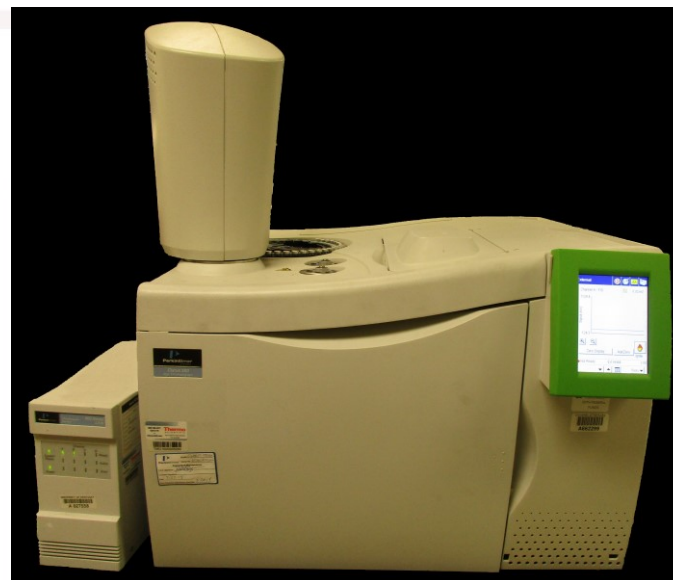


Δ⁸-Tetrahydrocannabinolic
Acid (Δ⁸-THCA)

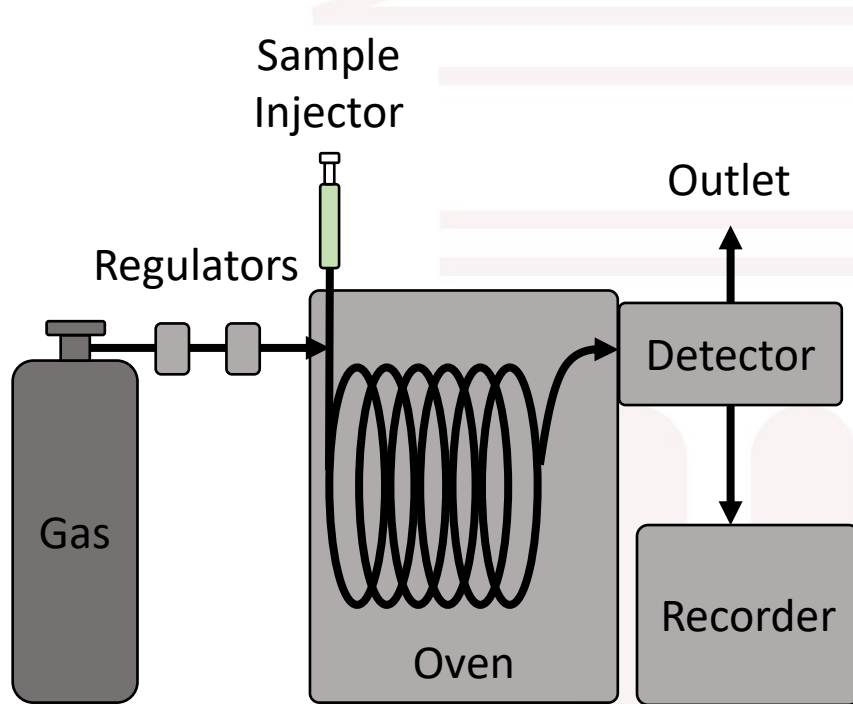
Quantification Methods

Liquid Chromatography

Gas Chromatography



GC Systems



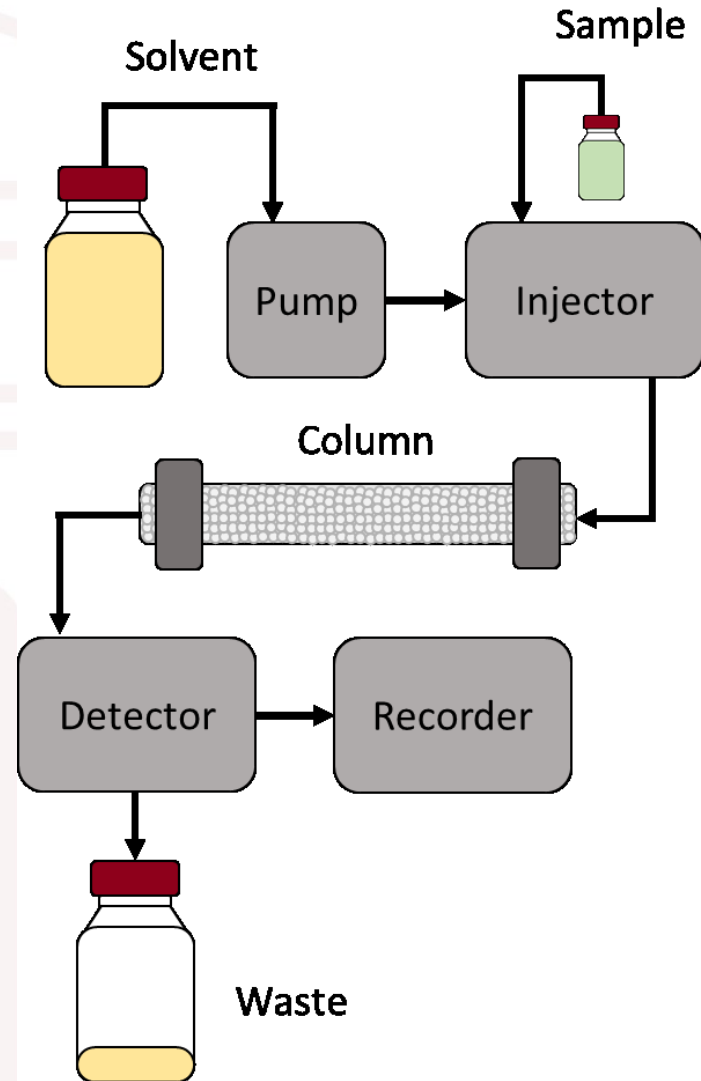
Gas Chromatography

- Heats samples until they volatilize into a gas
- Inherently decarboxylates cannabinoids
- Only quantifies total neutral cannabinoids

LC Systems

Liquid Chromatography

- High-pressure (HPLC) and Ultrahigh-pressure (UPLC) variants
- Uses a liquid flowing through a column to separate compounds
- Does not employ heat that decarboxylates cannabinoids
- Quantifies acidic and neutral cannabinoids separately





Detectors

GC Systems

Flame Ionization Detector (FID)

Mass Spectrometer (MS)

Others

HPLC Systems

UV Detector (UV)

Mass Spectrometer (MS)

Others

Sophisticated
physics to
quantify the
compound



Δ^9 -THC versus Total Δ^9 -THC

1. Δ^9 -THC Method = Δ^9 -THC
2. Total Δ^9 -THC = Δ^9 -THC + $0.877 * (\Delta^9$ -THCA)



Understanding Total-THC

- Some people argue THCA and THC are different molecules
 - technically true
 - closely related
- THCA easily converts to THC (the regulated intoxicant)

Hypothetical Example

Hemp?

Initial test results:

THCA: 6.2%
THC: 0.3%

Heat or Smoke



New test results:

THCA: 0.0%
THC: 5.7%

Hemp? NO!
Intoxicating?
YES!



Ratio of THCA to THC

$$\text{Total } \Delta^9\text{-THC} = \Delta^9\text{-THC} + 0.877 * (\Delta^9\text{-THCA})$$



Molecular Weight:

THCA = 358 g/mol

THC = 314 g/mol

$$\begin{aligned} \text{THC (g)} &= 1 \text{ g THCA} \times \frac{1 \text{ mole THCA}}{358 \text{ g THCA}} \times \frac{1 \text{ mole THC}}{1 \text{ mole THCA}} \times \frac{314 \text{ g THC}}{1 \text{ mole THC}} \\ &= 0.877 \text{ g THC} \end{aligned}$$



Regulatory Compliance

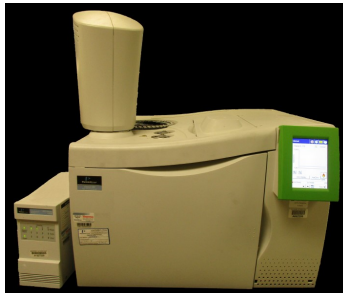
- USDA and Ohio both use the Total THC measurement system
- When quantifying compounds there is a level of uncertainty associated with the metabolite quantification process (“Measurement of Uncertainty”, MU)
- “Acceptable THC level” means a measurement of THC where the MU spans the 0.3% level or less

Provides Flexibility for Growers

“Acceptable THC” Examples

Example 1. Your crop comes back testing 0.25% THC ✓

Example 2. Your crop comes back testing 0.33% THC



Margin of error for the testing process is 0.04%

$0.33\% - 0.04\% = 0.29\%$
(Spans 0.3% value) ✓

Example 3. Your crop comes back testing 0.36% THC

$0.36\% - 0.04\% = 0.32\%$
(Does NOT span 0.3% value) ✗



Additional Testing

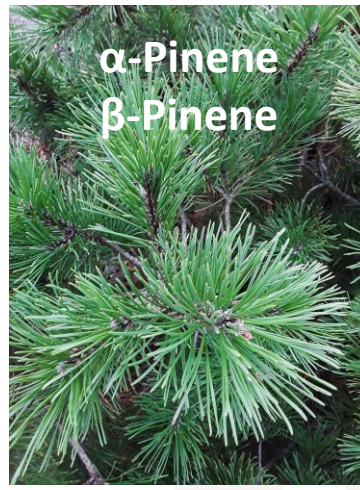
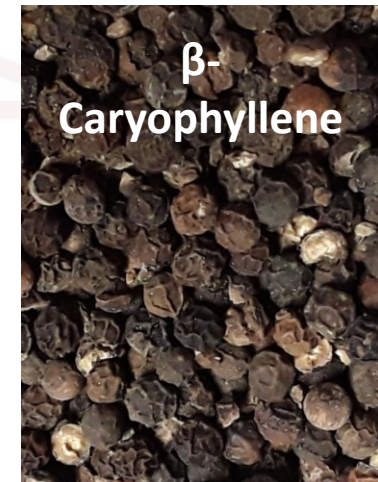
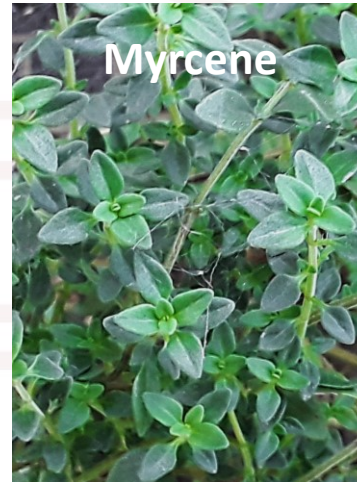
Only THC testing is mandatory for hemp

Optional tests include:

- Other cannabinoids
- Terpenes
- Moisture Content
- Foreign Matter
- Microbial Contaminants
- Mycotoxins
- Pesticide Residues
- Fertilizer Residue
- Heavy Metals
- Residual Solvents

Terpenes

- Primary flavoring/fragrance compounds of Cannabis
- Commonly analyzed in reports



Certificate of Analysis

Example

Chem Dawg CBD
Plant, Flower - Cured
Harvest Process Lot: METRC Batch: METRC Sample:

Analyte	LOQ mg/g	Mass %	Mass mg/g
THCa		0.51	5.1
Δ ⁹ -THC		0.19	1.9
THCV		0.02	0.2
CBDa		16.94	169.4
CBD		2.03	20.3
CBDV		0.05	0.5
CBN		<LOQ	<LOQ
CBGa		1.06	10.6
CBG		0.23	2.3
CBC		0.20	2.0
Total		21.24	212.4

$$\text{Total } \Delta^9\text{-THC} = 0.19 + 0.877 \times (0.51) = 0.63$$

Δ⁹-THC

Total CBD** (Calculated
Decarboxylated Potential)

21.24%

8.5%

CBGa	1.06	10.6
CBG	0.23	2.3
CBC	0.20	2.0
Total	21.24	212.4

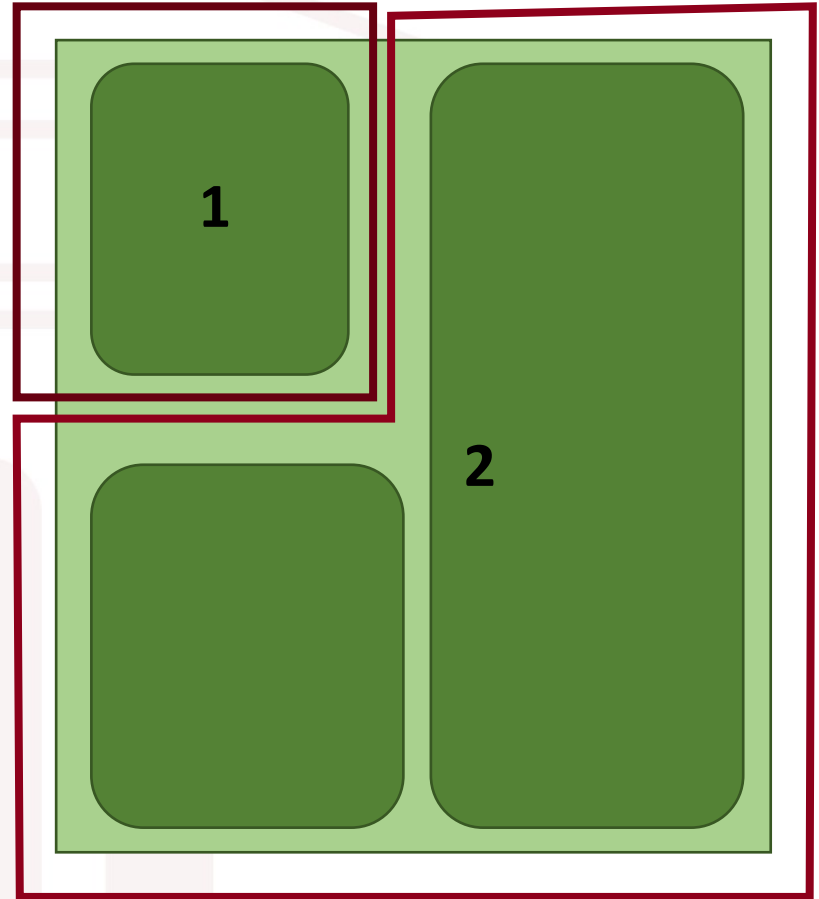


Testing Laboratories

- The Ohio Department of Agriculture (ODA) is the only official lab for testing THC for crop compliance
- ODA may contract with other labs for testing on their behalf if necessary
- Compliance testing only covers Total THC
- Growers should utilize private labs to monitor THC levels in their crops
 - especially important for metabolite crops

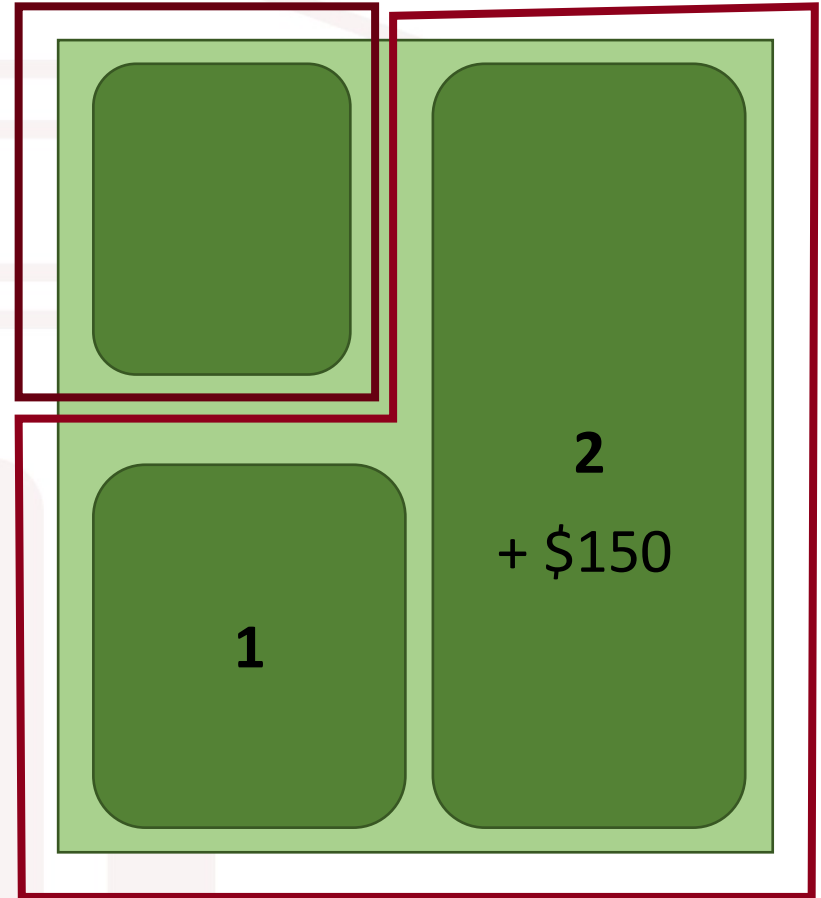
Growing Locations

- Growing location is a contiguous area (not broken by fences, waterways, tree lines, building walls, etc.)
- Each variety/strain at growing location requires separate testing



Defining a Sample

- Fees cover one test sample per growing location
- Additional tests are \$150 for preharvest samples





Testing Process

If crop is not harvested within 15 days of sampling another test for THC is required

SUNDAY	MONDAY	TUESDAY	WEDNESDAY	THURSDAY	FRIDAY	SATURDAY
			Notify ODA of intent to harvest			
		1	2	3	4	5
6	7	8	9	10	11	12
13	ODA collects samples	15	16	Intended harvest date	18	19
20	21	22	23	24	25	26
27	28	Crop must be harvested	30			



Questions