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# Lab Project Outline

**Project statement:** To determine the amount of THC and CBD present in cannabis bud samples, obtained from the BC Cannabis Store. These results will be compared to labeled content, to determine accuracy. This will be achieved using the Agilent Technologies G530 Accurate Mass Q-TOF 1200 series liquid chromatography - mass spectroscopy (LC-MS) instrument, the conditions can be seen below in figure 1. The column used is an Agilent Eclipse plus C18 column with an internal diameter of 1.8  $\mu$ m and column width/length of 2.1 x 100 mm. The ionization source used for the mass spectrometer portion of the instrument is an ESI+ source. Ions are analyzed by a Q-TOF mass analyzer and detected by a sensitive electron multiplier detector.

#### Daily objectives:

Day 1 - November 2nd		
	Perform experimental procedure as seen below, obtain products. Stock standards will be provided. If time permits, data will be run and obtained on the LC-MS. <i>Potentially run data prior to</i> <i>November 9th break, to ensure accurate samples</i>	
Day 2 - November 8th	If time permits in day 1:	If time does NOT permit in day 1:
	Analyze analytes to ensure it was accurately obtained. Start writing lab reports and lab presentations.	Run and obtain data on the LC-MS, analyze data to ensure accurate analytes obtained
		Lab report and lab presentation will be written in lab if time permits, and if not, on our own time.

Table 1. Daily objectives for lab experiment

# List of chemicals, supplies and equipment needed

- LC-MS
- THC and CBD standards Kingsley will provide
- Ethanol depends on method used
- Solvents for LC-MS as seen in Figure 1. (Acetonitrile for LC-MS Isaac is checking)
- Cannabis samples (Purchased from the BC Cannabis Store)
  - High THC
  - High CBD
  - Regular THC
  - Regular CBD
- Vacuum filtration
- Mortar and pestle OR cannabis grinder (purchased from the BC Cannabis Store)

## Instrument details and settings

Table 2. Optimized Experimental Conditions				
VCap:	3500V	Flow Rate:	0.4 mL/min	
		Injection		
Fragmentor:	60	Size:	0.2 µL	
Gas		Column		
Temperature:	350°C	Temperature	50°C	
Drying Gas:	13 L/min	Solvent A:	74.7% 18 mΩ Water, 25% Methanol, 0.3% Formic Acid	
Nebulizer:	60 psig	Solvent B:	99.7% Acetonitrile, 0.3% Formic Acid	
Sheath Gas				
Temp.:	325°C	Elution		
Sheath Gas		Gradient	100% A to 100% B in 5 min, hold at 100% B for 5 min,	
Flow:	12 L/min		then change to 100% A and hold for 5 min.	

Figure 1. The instrumental parameters for the Agilent Technologies G530 Accurate Mass Q-TOF 1200 series liquid chromatography - mass spectroscopy (LC-MS) instrument.

\*NOTE: There is another experimental parameters Kingsley can provide if needed (can also use water and 0.3% formic acid for the LC-MS solvents)

## Procedure

- Stock solutions of THC and CBD will be provided
  - A 1000 ppm CBD stock solution will be prepared using powdered CDB and methanol into a 25 mL volumetric flask
  - The THC stock will be provided
- The standard solutions will be prepared in 5, 10, 15, 20 and 25 ppm concentrations of THC and CBD standard solutions
- The cannabis strains will be ground up using one of the methods described in the list of chemicals and equipment needed. The bud strains may need to be grinded and mashed in a mortar and pestle.

#### Method:

Ground up cannabis was stored at -20°C

- 1 g cannabis + 20 mL ethanol, agitate 20 min, paper/vacuum filtration
- 100 ml solvent, agitate 20 min, paper/vacuum filter
- could do charcoal filtration but optional
- Evaporate solvent in water bath ~98°C under stream of nitrogen gas
- Reconstitute residue w/ ethanol to 100 mL (idk what that is)

## References

Benefits of Ethanol Extraction Systems for Extracting Cannabis <u>https://www.cbgbiotech.com/blog/benefits-of-ethanol-extraction-systems</u>

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