



Developing methods for the rapid identification of heavy metals and microplastics in CBD oil

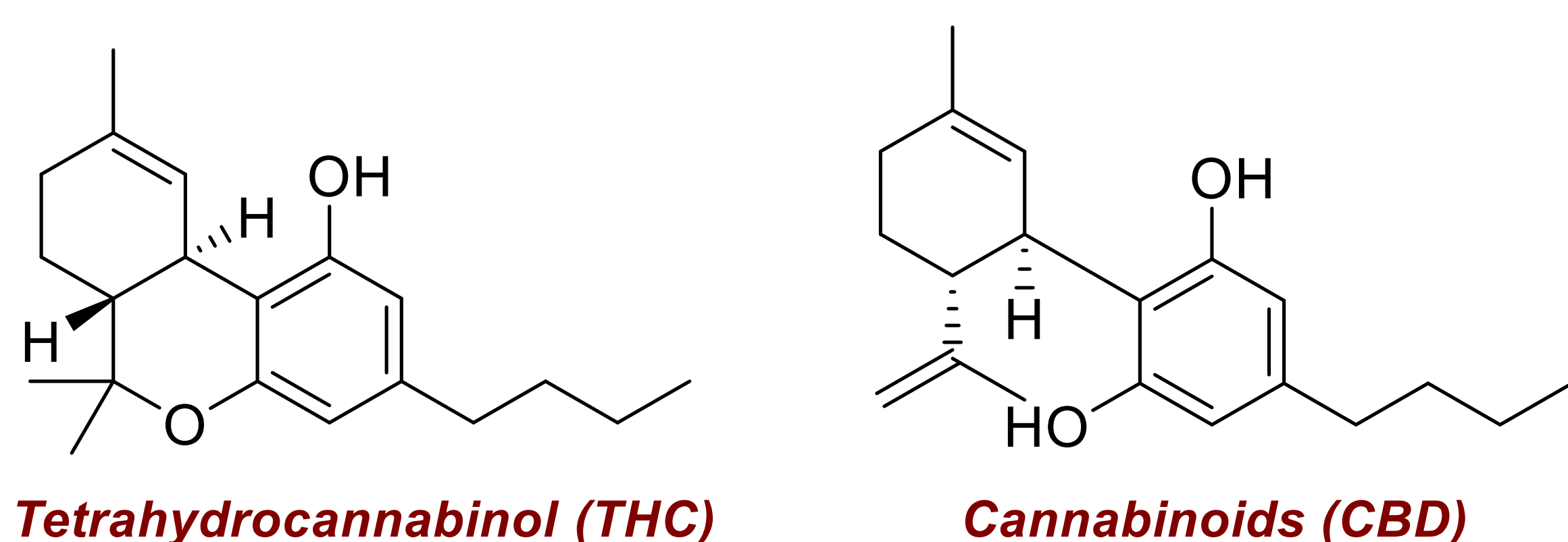
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Introduction

Cannabis has shown great promise for the treatment of many medical conditions. Tetrahydrocannabinol (THC) and Cannabidiol (CBD) are constitutional isomers with very similar chemistry (Scheme 1). The therapeutic application of cannabis and its constituent phytocannabinoids (i.e. THC and CBD), continues to garner significant clinical and public attention. There are, however, substantial uncertainties surrounding the nature and content of contaminants in cannabis plants. An in-depth understanding of plant contaminants and toxin effects on the stability of plant compounds and the effect on human health is necessary. The goal of this project was to develop reliable, rapid, efficient, inexpensive techniques for the determination of key contaminants within the cannabis plant and to accelerate research in this rising industry to ensure consumer/patient safety.



Scheme 1. Structures of THC (left) and CBD (right), two isomers as major constituent of cannabis plant.

The common cannabis contaminants include microbes, heavy metals, and pesticides. Their direct human toxicity is poorly quantified but include infection, carcinogenicity, reproductive and developmental impacts.¹ Microplastics are contaminants of concern; given their ubiquity they need to be regarded as a factor of global change.² Microplastics are defined as particles less than 5 mm, which are widely found in oceans and are also increasingly identified in freshwater. Although oceans and aquatic ecosystems have been the focus of microplastic contamination research for the last decade, microplastic effects in terrestrial ecosystems have recently moved into focus. To date, no quantitative data has been reported relating to cannabis microplastic contaminants.

Research Plan

Specific Aim 1: Development of a methodology to rapidly quantify the heavy metal contaminants, including arsenic, cadmium, cobalt, copper, lead and mercury in commercially available samples of CBD oil via Wavelength-Dispersive X-ray Fluorescence (WDXRF).

Specific Aim 2: Identification and mass determinations of microplastic polymers, including polyethylene (PE), polypropylene (PP), polyethylene terephthalate (PET), polyester (PES), polyamide (PA), polyvinylchloride (PVC), and polyurethane (PU) contaminants in commercially available samples of CBD oil using thermal analysis technique, viz., coupled Differential Scanning Calorimetry (DSC) and Thermal Gravimetric Analysis (TGA). The present study provides a cost-effective and straightforward method to determine the mass concentrations of the polymers by identifying the characteristic endothermic phase transition temperatures of the polymers in the samples.

Methods & Results

For Aim 1 all WDXRF analysis was made on a Rigaku Supermini200 instrument (Figure 1). All CBD oil samples were measured as liquids under a flow of helium gas. Each sample was analyzed for elements ranging from fluorine to uranium on the periodic table. Analysis showed that heavy metals were not present in the samples within the detection limits of the WDXRF instrument. However, analysis did identify the presence of trace amounts of silicon in at least 12 of the 25 samples measured.

For Aim 2 thermal analysis measurements were carried out on an in-house DSC 250 and TGA 550 from TA Instruments (Figure 1). The CBD oil samples were tested for the identification and mass determinations of microplastic (including polyethylene (PE), polypropylene (PP), polyethylene terephthalate (PET), polyester (PES), polyamide (PA), polyvinylchloride (PVC), and polyurethane (PU). First, the melting points of seven commercially-available plastic polymers (PE, PP, PET, PES, PA, PVC and PU) were obtained via DSC. Next, the oil samples were heated in TGA to 10 °C below the determined melting points of the polymers for 10 min in order to remove organic contaminants. Afterwards, the samples were tested by DSC for the detection of microplastics. Samples (5–10 mg) were loaded into sealed DSC aluminum pans and heated to 100 °C for 10 min to remove any water absorbed from the environment. The samples were then cooled to –50 °C and heated at a ramp rate of 2 °C/min to the determined melting points of the polymers. For each sample, ten scans were carried out to identify the correct phase transitions by observing three overlapping cycles. All measurements were carried out under a nitrogen atmosphere. Presumably, due to the low concentrations of microplastics in the samples, no characteristic endothermic phase transition temperatures were observed for the samples.



Figure 1. Rigaku Supermini200 WDXRF (top) and TA Instruments DSC 250 and TGA 550 (bottom).

Student Training

This project has supported the work of three undergraduate students who have been directly involved in each aspect of the research, including developing the analytical methods for the analysis of the CBD oil as well as data collection, and interpretation. The students are:

1) Ms. Isabella Riha is a senior BS Biochemistry major graduating in spring 2021. She has been accepted into the Chemistry Ph.D. program at Northwestern University beginning fall 2021 and recently was awarded an NSF-GRFP research fellowship.

2) Ms. Grace Anderson is a junior BS Biochemistry and BA Music double major graduating in spring 2022. She was awarded the prestigious Goldwater Scholarship, a nationally competitive STEM fellowship. She is FGCU's first recipient of this award.

3) Mr. David Siegel is senior BS Chemistry major graduating in spring 2021. He has been accepted to the Chemistry Ph.D. programs at UNC Chapel Hill, starting fall 2021.

Education Outcomes

We have developed a new course called the Chemistry of Medicinal Plants (CHM 2282C), which was approved by the State University System of Florida. The course will officially be part of the FGCU curriculum beginning in fall 2021 and will be incorporated into FGCU's Cannabis Studies Focus. Given that the official state level curriculum process takes over a year in the fall 2020 semester we offered the Chemistry of Medicinal Plants course in partnership with the FGCU Integrated Studies department under their IDS 3143 course number. In the fall 2020 semester, there were 33 students enrolled in the course.

Conclusions

Through our study we analyzed over two dozen commercially available CBD oil samples for the presence of toxic heavy metal and microplastic impurities. Our conclusions are that these contaminants are not present in the CBD oil samples we analyzed within the detection limits of our instruments. These findings should help reassure consumers and policy makers that CBD oil products are inherently safe. Additionally, this project facilitated the training of three undergraduate research students and the creation of a new course, Chemistry of Medicinal Plants (CHM 2282C).

Acknowledgments

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References

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2. Machado A. A. S., Kloas W., Zarfl C., Hempel S., Rillig M. C., Microplastics as an emerging threat to terrestrial ecosystems. *Global Change Biol.* 2018; 24: 1405–1416.