Using cannabis sativa for medicinal purposes presents a challenge to produce and deliver a consistent product. Product inconsistency is due to differences in cultivars and processing techniques that each has a significant impact on the resultant cannabinoid and terpene concentrations in the plant. Patients and consumers demand reliable and reproducible products, whether it be flower, edibles, or extracts. The industry depends upon a robust testing system to make decisions on dosing and content for the public's health and safety. This testing should include the assayed concentrations of major constituents and excipients in tinctures and extracts. It should also include moisture content, impurities, foreign organic matter, pesticides, microbial and fungal organisms, metals, and residual solvents.[i] Testing the whole plant or consumable products requires that samples be prepared to allow for the consistent analysis of the material. In the case of cannabis, the inflorescence is typically separated from the remainder of the plant because most of the components of interest are concentrated within the female flower. The initial step is homogenization through mechanical means or maceration into fine particles. This process must be done to not contaminate the sample with previous samples or leave residual sample in the equipment. For example, if resin from the flower preferentially adhered to the canister, it would yield lower concentrations of specific cannabinoids. This maceration can be done mechanically or combined with a

solvent at this stage or later as a separate step. Once solubilized, there are many extraction techniques. All take advantage of the relative lipophilicity of the cannabinoids by placing the sample in an organic solvent. Methanol and alcohol are commonly used but many other substrates can be used including some that have high toxicities that must in turn tested for. The excess solvent is removed, in the case of methanol by evaporation. The extract is then ready for separation and analysis of its constituent components.

Liquid chromatography is the most common technique for this separation. It is designated normal or reverse phased. In both types, there is a mobile and a stationary phase. In normal phase chromatography, the stable phase is hydrophilic, and the mobile phase is lipophilic. When analyzing cannabis, more commonly reverse phase high performance or ultra performance liquid chromatography is used.[ii] In reverse phase chromatography, the mobile phase is hydrophilic with the stationary phase lipophilic. Liquid CO2 extraction has become the preferred technique because of the low temperatures and the lack of residual solvents.

Alternatively, gas chromatography can be used as the mobile phase but has the limitation of creating heat that may decarboxylate the parent compound before it can be analyzed. Gas chromatography allows the separation of volatile constituents detected through either flame ionization (FID)or mass selective detectors.

In all techniques, the mobile phase extract is pumped or passed through a column at a controlled flow rate through a stationary medium, and the eluded components separate based upon their molecular weight and their relative affinity for the mobile phase or the stationary phase. The separated eluded components are analyzed by sensors as they emerge. They can be either quantified, or collected as purified components.

The extraction technique will significantly impact the relative quantities of THC and other metabolites and, therefore, must be consider

ed when analyzing the results.

To more precisely analyze the components of cannabis, most labs combine liquid or gas chromatography with mass spectrometry. H-mass spectrometry takes advantage of the spin of protons in molecules and the absorption of energy needed to move that proton out of alignment with an applied magnetic field. The resultant spectrograph produces spikes of relative intensity based on the quantity and relative shielding of protons related to neighboring carbon atoms and associated hydrogen atoms. The observed peaks on the graph will have frequency responses that can be reproduced and compared to established references.

No one technique can provide a complete analysis of cannabis products.

---

[i] Upton R, et al Ed's. Cannabis inflorescence (Cannabis app.) : Standards of Identify, Analysis, and Quality Control. Scott's Valley, CA: American Herbal Pharacopeia, ed;2013.

[ii] Fischedick,JT et al, A qualitative and quantitative HPTLC densitometry method for the analysis of cannabinoids in Cannabis sativa L Phytochem Anal: PCA. 2009;20(5): 412-426.