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Lesser known cannabinoids and terpenes may be helpful for patients. Here the author shows why, and how patients can choose more effective medical cannabis products.

Analysis of Veterinary Hemp-Based Oils for Product Integrity by LC/MS

Ben Nie, Jack Henion, and Joe Wakshlag

Some hemp-based products are mislabeled regarding their level of CBD. The author explains why and how that happens.

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Inter-Lab Variation in the Cannabis Industry, Part II: Solutions

In the first installment of this two-part series, I discussed the problem of inter-laboratory variation in the cannabis testing industry. In this second installment, I offer potential solutions to the problem. These include more and better state oversight, proper sample storage, industry agreement on standard methods, and orthogonal testing. Despite its prevalence, I believe the inter-laboratory variation problem is solvable.

Brian Smith

It is hard to believe that we are already up to the fifth installment of this column. To review, the previous columns introduced the scope and purpose of the series (1), discussed error, precision, and accuracy (2), covered the problem of inhomogeneous samples (3), and in the last column I shared with the industry my thoughts on the problem of inter-laboratory variation and its causes (4). Here is that list of the causes as I see them:

1. Lack of standard methods
2. Sample inhomogeneity
3. Lack of appropriate standards
4. Sample preparation variability
5. Sample instability
6. Large factor dilutions
7. Treating analytical methods as intellectual property
8. Human error

It’s a long list and the cause of a complex and knotty problem. I have been a practicing analytical chemist for four decades, and been intimately involved in cannabis analysis since 2014, when it was first legal to do so. Therefore, I believe I am uniquely qualified to take a step back, analyze the problems of the cannabis analysis industry, and suggest solutions. I do this not out of malice but love. The newly opened field of cannabis science is like an unexplored continent, where every day fascinating new discoveries are being made that might benefit mankind. For those discoveries to bear fruit, the industry must be based on science and professionalism. These solutions then are offered in the hope of making a small contribution to this goal. Each solution detailed below will address one or more of the causes of the inter-laboratory variation problem discussed in the last column.

Potential Solutions

The ultimate solution to many of the causes of the cannabis laboratory inter-laboratory variation problem is legalization of marijuana at the federal level in the United States. This would allow cannabis scientists to come out from the shadows, pursue all research avenues, and feel free to share their work. It would allow the U.S. Food and Drug Administration (FDA) to become involved in the regulation of the manufacturing and analysis of cannabis medicines, and it would give a green light to the National Institute of Standards and Technology (NIST) to issue appropriate cannabis standard reference materials (SRM).

However, until that happy day comes, we as an industry will have to struggle with and try to solve this problem on our own. I believe that if we can make progress it will raise public confidence in our ability to make safe and effective medicines, increasing the likelihood of expanded legalization. Meanwhile, here are steps we can take now to start solving the problem.

Improved State Oversight

As of now it is state governments that bear the brunt of regulating the cannabis industry. The fact that the inter-laboratory variation problem exists in multiple states means that no one state government has completely figured out how to regulate cannabis testing properly. Wouldn’t it be great if state regulators and cannabis scientists from around the country got together, harmonized regulations, and agreed on best practices for cannabis analysis? In the absence of federal oversight, it would go a long way towards solving this problem. Until then, individual states could begin by choosing a “golden laboratory” within the state whose methods are trusted enough that it would be the go-to laboratory for any questions about samples analyzed elsewhere. As I have pointed out previously (4), the lack of SRMS
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in our industry means that true accuracy, or knowing how far away you are from the true value, is impossible to achieve. In this absence, it makes sense to pick a laboratory whose instrument, personnel, and method have been properly vetted and trusted enough to put a stake in a ground and be the "true value" that all other laboratories in a state must compare their results to. Alternatively, state regulators could be more specific in their regulations, calling out specific extraction methods for example that are used industry-wide and known to work.

State personnel should also perform blind round-robin studies, similar to those already published in the literature (5-7), by sending the same sample to multiple labs, tabulating and comparing the results, and publishing them so the public knows who is and who isn’t doing a good job. To be clear, the inter-laboratory variation problem has been observed with cannabis laboratories that are state and International Organizational for Standardization (ISO) licensed and certified (5).

This means these certifications by themselves are not sufficient to ensure every laboratory in a state gets similar results on the same sample. This indicates that state governments need to step up their game to solve this problem.

**Industry Organizations Need to Speed Up Their Work**

At the moment the United States Pharmacopeia (USP), for Standardization (ISO) licensed and certified (5).

State laboratories need to do this. For example, state regulators could be more specific in their regulations, calling out specific extraction methods for example that are used industry-wide and known to work.

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**Representative Sampling**

Briefly, given the inhomogeneity of cannabis samples, particularly plant material, the taking of one aliquot, testing it, and reporting out one set of results is inappropriate. Statisticians tell us that the best way to overcome inhomogeneity is to test multiple aliquots and average the results (3).

I realize that testing multiple aliquots is time-consuming and expensive, but it is the only way to obtain representative data on our samples and begin to chip away at the problem of inter-laboratory variation.

Perhaps state regulators can step in here and require more than one aliquot of each sample be tested, particularly for failed samples or samples close to action limits. Also, laboratories need to consider methods other than chromatography such as infrared spectroscopy (7) that are faster, cheaper, easier, and more readily lend themselves to analyzing multiple aliquots.

**Analyzing Samples Quickly**

Work in the literature shows that cannabis samples are unstable, some with half-life of just eight months (8-12). Thus, these samples should be analyzed as soon as possible after collection. This means cannabis laboratory clients need to rush samples to the laboratory, and laboratories need to turn around samples as quickly as possible. I realize many laboratories may be swamped with samples, and five-day or even seven-day turn-around times may be common. But this needs to be the exception rather than the rule. The industry needs to invest in the personnel and equipment to speed up analyses so samples are analyzed while they are fresh.

**Proper Sample Storage**

Since we know cannabis samples are unstable, and until we have further data on ideal storage conditions, all samples collected should be stored in a refrigerator in the dark until they are needed for analysis. State regulators should insist upon this.

**Orthogonal Testing**

Orthogonal testing means using a second analytical method as a referee for a primary method. For example, it has been shown that mid-infrared spectroscopy can accurately determine cannabinoid and terpene profiles in cannabis distillates and extracts (5,12,13). Side-by-side studies on the same sample set can be used as a sanity check on high performance liquid chromatography (HPLC) potency methods. If the HPLC suddenly starts disagreeing with the second method, it may indicate that some undetected change in the HPLC method has occurred that needs to be investigated. Some laboratories are already doing this (14).

**More and Better Training**

At the end of the day our analyses are only as good as the quality of the people performing them. The existence of the inter-laboratory variability problem and my...
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own observations and experience prove to me that we as an industry need to improve the quality of our analysts. This means cannabis laboratories need to invest more time in training their current employees, and hire employees with appropriate degree levels and experience.

To draw the analogy to the pharmaceutical industry, laboratory managers and directors there typically have graduate degrees in analytical chemistry and many years of experience. Given that cannabis is medicine and should be tested like medicine, I strongly feel that laboratory managers and directors in the cannabis industry should have a graduate degree in analytical chemistry and significant experience. Technicians should have appropriate degrees and extensive on-the-job training.

This is another place where state regulators can step up, requiring continuing education for current laboratory employees, and insisting new hires have appropriate educational levels and experiences.

Conclusions

The inter-laboratory variation problem in the cannabis analysis industry is real and threatens the health and safety of consumers and the future of our industry. It has a number of causes, including a lack of standard methods and reference materials. These problems are exacerbated by the lack of marijuana legality at the federal level in the United States. However, there are things the cannabis industry can do now of its own accord to ameliorate the problem including agreeing on standard methods, representative sampling, proper sample storage, orthogonal testing, and more and better training of laboratory personnel. Only if we as an industry acknowledge this problem and begin working on solutions now will our desired goal of widespread legalization ever occur.

References

4) B.C. Smith, Cannabis Science and Technology 2(2), 12-17 (2019).
14) J. Strull, private communications.

About the Columnist

Brian C. Smith, PhD.
is Founder, CEO and Chief Technical Officer of Big Sur Scientific in Capitola, California. Dr. Smith has more than 40 years of experience as an industrial analytical chemist having worked for such companies as Xeros, IBM, Waters Associates, and Princeton Instruments. For 20 years he ran Spectros Associates, an analytical chemistry training and consulting firm where he improved their chemical analyses. Dr. Smith has written three books on infrared spectroscopy, and earned a PhD in physical chemistry from Dartmouth College.
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Understanding the Science of Cannabis Product Development

The rapidly expanding global market for cannabis products should be based on the medical science of cannabis and how it interacts with the human endocannabinoid system. Companies that base their brands on the science of the entourage effect and develop products around plant genetics using cutting-edge extraction technology will rule global market share, reshape our medical system and recreate how we choose to find relaxation and stimulation. Current political and economic situations have created non-science-based markets and products that are not serving medical patients, and are not designed to fill the research gaps we desperately need to fill to legitimize the emerging anecdotal evidence. By understanding whole-plant extractions better, allowing for their medical use and removing restrictions on research and development, businesses will be able to better plan for long-term profitability and sustainability.

Amber Wise and A.C. Braddock

Recorded history has been written by the victors, and the propagandizing of Cannabis sativa L. in the U.S. is an obvious and tragic example. We quickly forget the history on which we should be making decisions and worse, we sometimes base our future on a revised version of it. Less than 100 years ago, a handful of men took thousands of years of knowledge and successfully distorted it for their own financial and political gain. We allowed the copacetic relationship between cannabis and humans to be severely maligned by propaganda, and hundreds of thousands of lives have been medically, socially and financially destroyed to serve a few (1). While underground medical practitioners, caregivers, and scientists have made some progress in unraveling the last century of lies, we are still on the path that a small group of men devised (2). Current progress to legalize cannabis will further our understanding of this important plant as medicine, but education and de-stigmatization on a larger scale needs to occur.

Traditionally, medicines have been created from plants and were often whole-plant preparations or basic tinctures or extractions until science advanced and we understood how to better isolate the most active agent in plants. There were many known preparations of Cannabis sativa and it was used widely for treating a number of ailments, primarily pain management. One of the first medical isolations was the alkaloid morphine from opium poppies by Friedrich Wilhelm Adam Sertturner in 1805 (3). This was closely followed by other isolates as well as the invention of the hypodermic needle in the early 1850s. Since cannabis is not water soluble, it could not be directly injected into people, which contributed to its medical decline. Its use was completely decimated after its political damning in the early 1900s (4). Morphone and other opiate-derived drugs quickly supplanted cannabis as the primary pain management tool. From there, chemists learned to synthesize morphine, and its derivatives have been developed into multiple products, many of which have now devastated millions of people’s lives.

Aside from the water insolubility and political pressure, another issue facing cannabis was determining a “safe” dosage. Once its use and research declined, however, it’s difficult to argue that the creation of the “one-pill-fits-all” system that developed in pharma has been safe or effective for everyone when one considers the addiction, side effects, and deaths associated with prescription drug use. We have also recently discovered that people have genetic predispositions, as well as metabolic differences, to drug toxicity and efficacy (5,6). This new area of research in individual reactions and efficacy is very complicated, and requires extensive testing, to pair an individual with a drug that will be effective and not have harmful side effects.

Now, more than a century later, the pharmaceutical industry, based on isolation of single molecules, has created a clinical trial process that is not equipped to handle the multitude of compounds and natural variations of whole-plant preparations. This leaves us in a similar position as in years ago of having a lack of the “approved” type of data that other drugs were allowed to explore because of their federal scheduling. However, the rising tide of anecdotal evidence of overwhelmingly safe cannabis use as a medicine is piling up, and medical doctors and researchers are starting to piece together some of the important components for specific treatments and preparations.

The safety of using whole plant applications in anecdotal evidence, as well as its historical use, lends itself to researching chemovars to support more immediate access for medical treatments.
Isolation is laborious and expensive in practicable terms for commercial production of products. The only way to procure a single pure molecule is by using preparatory chromatography after extraction. This process is not scalable for the production required to extract the low quantities of cannabinoids (CBD) found in hemp. One way to alleviate this issue is to grow cannabis for high levels of a specific cannabinoid and extract for its boiling point. If we do not find a way to approve whole plant research and use, this industry will be taken over by the usual synthesizing of the molecules to produce quantity.

Medical cannabis legalization in the U.S. began in 1996 when California legislatively passed Proposition 215 (7); from there Oregon, Washington and Alaska all became legal medical use states. They legalized on the medical use of Cannabis sativa L.—the whole plant and not a low tetrahydrocannabinol (THC) varietal recently defined as “hemp.” There were no restrictions on cannabis cultivars, and there were no recommended dosages. A patient could simply grow and extract the medicine they needed from the plant and treat themselves like humans have done for thousands of years. To date, no death has occurred from cannabis consumption, no prevalent addiction exists and the plant has proven to help reduce the need to pile pharmaceutical drug on top of pharmaceutical drug to treat what are often debilitating side effects. We knew as early as the late 1880s that cannabis extracts could be successfully used to wean people off opiate addiction and yet we continue to ignore the science because of the politics.

In 1889, an article by Dr. E. A. Birch in The Lancet, still one of the world’s leading medical journals, outlined the application of cannabis for the treatment of opium and chloral hydrate withdrawal symptoms—the mixture reduced the opium craving and acted as an anti-emetic, which is a drug that is effective against vomiting and nausea.

So how can we understand some of this history through the lens of extraction and formulation science? How do we avoid repeating history’s mistakes when we think about new product development? How can we incorporate modern scientific advancements with traditional knowledge? One of the most significant and recent scientific discoveries since that time is the endocannabinoid system (Figure 1) (12). This internal system of connected signaling mechanisms regulates a huge variety of effects in our bodies: metabolism, homeostasis, restfulness

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and anxiety, pain, sexual health, mood and more. Our bodies naturally have molecules that interact with our endocannabinoid system, and there are other molecules (such as THC and other cannabinoids) that also interact with the same receptors. Additionally, the evidence is accumulating that having more of the other naturally occurring biomolecules from the cannabis plant, such as terpenes and flavonoids, elicits very different effects that work far better than single molecule preparations alone. This theory has been named the entourage effect, and, given the complexity and number of different compounds in cannabis, it will take some time and careful research to fully understand the various important active compounds for treating specific ailments (13). There are clearly specific cultivars with specific chemical profiles that can be grown reproducibly that are useful for specific medical treatments, and not for others. But it’s only a matter of time before the analytical chemistry and big data analyses can begin to make the connections. This is where understanding the complexities of extractions, isolations and formulations will be useful to harness the full capability of the plant.

How do we plan science-based formulations in a constantly and rapidly changing regulatory environment? For a business plan to be of value in this growing industry, its 5 to 10 year plan must be considered in this mercurial and changing environment. As an example, many startups have moved from “cannabis” to “hemp” when creating CBD product-based businesses (9). This migration hasn’t been based on science but simply because the regulations around cannabis-derived CBD are severely restrictive, legally punitive and financially unsupported by a federal banking system. In fact, the science shows over and over again that isolating and purifying the cannabinoids vastly reduces their medical efficacy.

Cannabis is an extremely malleable plant that has the ability to be cultivated for high levels of certain cannabinoids. It seems strange that current regulations focus on one or maybe two specific molecules (THC and CBD) that don’t even naturally occur in a freshly harvested cannabis plant. Why is it that the legalization platform did not occur on cannabigerolic acid (CBGA) or cannabigerol (CBG)? As the mother of all cannabinoids (the molecular precursor for the enzymatic processes to synthesize the other naturally occurring cannabinoids), CBGA is not psychoactive and therefore could have been politically acceptable (8). With the growing ability to genetically modify plants, we have begun to breed for the end-product, which can improve processing and medical efficacy through more full spectrum profiles.

Because of political bias, we are growing an industry based on a single molecule—CBD—that has a medical Bell Curve. The Bell Curve refers to medical treatment and sustained efficacy. In a Bell Curve response, over time, an increase in the dosage does not correspond to an increase in efficacy. However, when a whole plant treatment is provided, efficacy increases—this is medicine (10). Unfortunately, legislators and regulators did not and largely still do not know this important fact when building a legal foundation for an industry based on medical use. Our challenge as scientists, and business professionals, is to inform legislators, regulators and the public that single-molecule products are being produced because of our political environment, not because it is rooted in medical science. Unless the medical science is based in research around the newly discovered endocannabinoid system, we have not progressed past the disinformation spread by prohibition and 19th century medical practices.

I have had the privilege to read and watch a multitude of business pitches in the past few years. What is truly disturbing are two things. First, CBD-only products are being touted as a “cure all.” CBD is a powerful cannabinoid, but it is not a “cure all.” Second, the laser focus on only CBD and the continued demonizing of THC (the so-called “recreational cannabinoid”) is continuing to support and spread the propaganda around Cannabis sativa L that began in the early 1930s. Not only has CBD been shown to work better when there is a small amount of THC present (and most often other terpenoids and biomolecules—the entourage effect), but THC itself is medically relevant for a wide range of ailments.

Figure 1: The human endocannabinoid system, an internal system of connected signaling mechanisms, regulates a huge variety of effects in our bodies: metabolism, homeostasis, restfulness and anxiety, pain, mood and more (12).
The sheer number of companies adding CBD into everything is damaging the cannabis industry. Not only are companies acquiring CBD from questionable sources that contain impurities and pesticides, they are then adding this into products that are often the only legal form of medicine for patients to consume. They are marketing CBD products that contain miniscule or no actual CBD in them. This is snake oil and it won’t take long for people who try it to make the correct conclusion that these CBD products do not work. The danger is they will assume all CBD products are fake and will stop buying all medically active cannabis products. Again, science is not prevailing here but instead political restrictions and opportunistic product developers.

Along these lines, the descriptors around whole plant extracts need to be defined. I read one pitch deck that naïvely bragged their products would be “full-spectrum distillates.” The distillation process is designed to purify mixtures and separate molecules from those with different boiling points and molecular weights. The vast majority of all the other biomolecules such as...
terpenoids, fats, lipids, chlorophyll and waxes are removed. Therefore, by definition, a distillate cannot be “full spectrum”(11). To produce a full spectrum extract for pharmaceutical grade products, the most commonly used methods are supercritical CO₂ and ethanol (14) (Figure 2). Supercritical and subcritical CO₂ (Figure 3) create a vast variety of end products that can contain precious terpenes, and be tuned to acquire targeted cannabinoids in the material (15). A warm ethanol extraction is by far the most efficient method, and will perform an exhaustive extraction, which means pulling all of the compounds out of the plant, including chlorophyll that can be quickly filtered from the crude oil. The down side is the loss of terpenes in the solvent recovery process. But a preprocessing of steam distilling will remove purified terpenes that are added into a final formulation can create a more truly full spectrum extract. Cold ethanol extraction will also help retain terpenes. It is a much slower process and not as efficient, but it can eliminate the need for post processing the crude oil.

Understanding how different extraction protocols are performed and the various outcomes is crucial in the product development process. A combination of extraction methods are often successfully used to create greater processing efficiencies and a greater diversity of products on the shelf with a single batch of material. However, it’s not only a good extraction and formulation that will make a successful product or brand. Every product line is dependent on a message and the line should be created to solve a problem or fill a niche. In the educated connoisseur and medical markets, how and why it is made is the inherent value of the brand. Consumers will pay more for products and brands that bring value to their community. These are the consumers that are aware of the social destruction and medical ills created and propagated by prohibitionists, and will listen when you create a product that restores the value and honor of the relationship between humans and cannabis.

The scientific and business leaders of this growing industry cannot perpetuate the disinformation of the past. We cannot build an industry based in health and well-being and sustain it without bringing the public and politicians along with us into an era of new medical science based on the endocannabinoid system, the entourage effect, and how to create whole-plant formulations and their application to individuals. It is time we investigated the real science, and develop products for brands that take our culture to the next step, and not pander to old medical practice, the traditional pharmaceutical model and short-sighted investors. It will take a concerted effort. Product development and placement are the platform on which to build and heal our bodies, minds, culture and environment.

References
4) https://www.pbs.org/wgbh/pages/frontline/shows/dope/etc/cron.html
7) https://ballotpedia.org/California_Proposition_215_the_Medical_Marijuana_Initiative_(1996)

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Beyond Potency: The Importance of Terpenes

Cannabis analysis over the past decade has progressed from non-existent idea to an evolving daily practice. The initial targets for analysis were the cannabinoids in order to determine the psychoactive compound concentrations in the cannabis products. Now, the field is expanding to encompass other important target compounds and elements. Certainly, compounds like pesticides and elements such as heavy metals are of particular importance to health and safety of consumer products. But, there are thousands of other potential analytical targets in cannabis which are still relatively unexplored. These compounds contribute to the health, nutritional, and more esoteric aspects of cannabis such as flavor, scent and overall composition.

Patricia Atkins

Many chemical compounds can be responsible for scent and flavor in botanicals. Taste and smell are two of our primary sensory systems in which humans perceive the world. Both taste and smell are chemoreceptive senses meaning that there are specialized sensory receptor cells that convert a chemical substance to a signal such as neurotransmitter or an action potential in a nerve cell. There are two types of chemoreceptors: distance and direct. Distance chemoreceptors are present in the olfactory system (smell) and allow the detection of chemicals in a vapor or gaseous state. These receptors allow for the detection of odors and pheromones. Direct chemoreceptors are present in the gustatory system (taste).

Chemical compounds interact with chemoreceptors in the mouth or nose and create a response. In human beings, the tongue is the most important sensory organ for taste but there are chemoreceptors all over the mouth. Salivary glands in the mouth produce saliva which is a chemical cocktail of water, electrolytes, cells and enzymes. The saliva is also a liquid or aqueous matrix for the food to interact with the taste buds as well as begin digestion of starches and fats. Without the assistance of saliva, the taste buds would not be able to chemically interact fully with the various flavors of food.

The chemoreceptors of the tongue and nose are primarily G protein-coupled receptors (GPCR). In addition to the GPCRs, the tongue also contains channels. GPCRs in the mouth are proteins that bind to ligands and begin signaling action potentials within the brain to differentiate between the three of the five basic tastes: sweet, bitter, and umami (savory). The taste of sour and salty are perceived through ion channels which are pores in membrane proteins that allow ions to pass into cells and through membranes. These chemoreceptors are located in taste buds around the mouth and on the tongue.

Each taste is triggered by different groups of compounds and different mechanisms of action. (See Table I). Sweet tastes are triggered by compounds such as carbohydrates and carbonyls activating GPCRs on the front section of the tongue, while salty tastes are triggered by alkali metals via ion channels set just behind the sweet areas of the tongue. These chemical reactions are transformed into neural impulses and travel along the various facial and major nerves to centers in the brain which then interpret the impulses and create taste perception. Impulses sent to the somatosensory and frontal cortex of the brain are perceived as a conscious understanding of taste, while impulses sent to the amygdala and hypothalamus perceive an emotional context of taste. Finally, the hippocampus gives us the memories of taste. These perceptions of taste along with the mouth feel of food or texture, the olfactory impulses associated with smell and the sensation associated with temperature, pain and pressure (chemesthesis) combine to create the impressions of flavor.

Chemicals which produce flavor or fragrance contain functional groups which activate the corresponding taste receptors to perceive taste which combine with the other senses to produce familiar flavors. The most common functional group in flavors is carbonyls such as esters, aldehydes and ketones, etc. Other groups which produce flavors are carbohydrates, acids, salts, proteins and terpenes (See Table III). Terpene is the common term for a large group of compounds that contribute to flavor
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and smell of botanical products. Isoprene or 2-methyl-1,3-butadiene (See Figure 1) and its polymers is the main base of natural rubber and the structural base for terpenes and terpenoids, even though isoprene is not part of the reactions which produce terpenes.

The actual mechanisms for the synthesis of terpenes are derived from units of isopentenyl pyrophosphate (Figure 2). The two metabolic pathways to synthesize terpenes are the Mevalonic acid pathway (MVA) or the MEP/DOXP pathway. The pathways are usually exclusive to the type of organism with green algae producing terpenes via the MEP pathway; humans and fungi via the MVA pathway and plants producing terpenes from both pathways (1). Terpenes and terpenoids can be subdivided into smaller groups by the number of carbon atoms or by the number of isoprene base units the compound contains (Table II).

Monoterpenes are lower-molecular-weight terpenes and are responsible for lighter floral fragrances. These lighter-weight terpenes can volatilize quickly after harvest and during processing involving heat and decarboxylation. Sesquiterpenes are larger-molecular-weight terpenes and have a heavier fragrance such as sandalwood or musk, volatilize at higher temperatures, and remain after many processing steps.

Many consumer products including perfume, flavorings, wine and beer are dependent upon terpenes for the character, flavor and fragrances they impart upon products. The floral notes of wine grapes can be traced back to terpenoids such as Damascene and Geraniol, which are also found in roses. The diversity of terpenes and terpenoids are recognized by all the range of scents and flavors they produce from the pine scent of pinene (the most widely encountered terpene in nature) to the lavender and mint notes associated with linalool. The flavor and aroma of hops are critical to beer, especially myrcene, β-pinene, β-caryophyllene and α-humulene.

Many applications for the use of cannabinoids for health benefits have been published over the past decade. There have also been numerous publications of the health effects of various terpenes and terpenoids which suggest

### Table II: Terpene groups and examples

<table>
<thead>
<tr>
<th>Terpene Group</th>
<th># Isoprene units</th>
<th># Carbons</th>
<th>Terpene Example</th>
<th>Terpenoid Example</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemiterpene</td>
<td>1</td>
<td>5</td>
<td>Isoprene</td>
<td>Isovaleric acid</td>
<td>Isoprene is the only hemiterpene.</td>
</tr>
<tr>
<td>Monoterpenes</td>
<td>2</td>
<td>10</td>
<td>Limonene</td>
<td>Terpineol</td>
<td>Large group of volatile and semi-volatile compounds</td>
</tr>
<tr>
<td>Sesquiterpene</td>
<td>3</td>
<td>15</td>
<td>Humulene</td>
<td>Farnesol</td>
<td>Large group of volatile and semi-volatile compounds</td>
</tr>
<tr>
<td>Diterpenes</td>
<td>4</td>
<td>20</td>
<td>Taxadiene</td>
<td>Cafestol</td>
<td>Precursor compounds for production of retinol and retinal</td>
</tr>
<tr>
<td>Sesterterpenes</td>
<td>5</td>
<td>25</td>
<td>Ophiobolin A</td>
<td>Geranylnerolidol</td>
<td>Rare group of terpenes mostly from marine sources</td>
</tr>
<tr>
<td>Triterpenes</td>
<td>6</td>
<td>30</td>
<td>Squalene</td>
<td>Sterols</td>
<td>Squalene is shark liver oil and the precursor to some steroids</td>
</tr>
<tr>
<td>Sesquiterpenes</td>
<td>7</td>
<td>35</td>
<td>Tetraprenylcurcumene</td>
<td>Ferrugicadiol</td>
<td>Mostly produced by microbes</td>
</tr>
<tr>
<td>Tetraterpenes</td>
<td>8</td>
<td>40</td>
<td>Lycopene</td>
<td>Caretenoids</td>
<td>Family also includes carotenoids</td>
</tr>
</tbody>
</table>

### Table I: Compounds and mechanisms of tastes

<table>
<thead>
<tr>
<th>Taste</th>
<th>Compounds</th>
<th>Mechanism</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sweet</td>
<td>Proteins, Carbohydrates, Carboxyls</td>
<td>G-Protein Receptors</td>
</tr>
<tr>
<td>Salty</td>
<td>Alkali Metals, Na+, K+, Fe+</td>
<td>Ion Channels</td>
</tr>
<tr>
<td>Sour</td>
<td>Acids</td>
<td>Ion Channels</td>
</tr>
<tr>
<td>Bitter</td>
<td>Phenols, Catechins, EGCG</td>
<td>Type-2 Taste Receptors</td>
</tr>
<tr>
<td>Umami</td>
<td>Amino acids, Nucleotides, Caffeine</td>
<td>Variant G-Protein Receptors</td>
</tr>
</tbody>
</table>
terpenes are often anti-inflammatory and anti-oxidants. (2,3). Cannabis is rich in both cannabinoids and terpenoids, which research suggests can have therapeutic and medical value. The most common terpenes found in cannabis variants are myrcene, caryophyllene and many others (see Table III). Current research is investigating the possible synergistic effects of cannabinoids and terpenes. It is often well-accepted that there are synergistic effects between bioactive compounds and nutrients in foods and supplements and not just a single compound (4). Research has suggested the presence of terpenes in cannabis products can alter the pharmacokinetics of cannabinoids (5,6). There are still questions being researched as to which cannabis product could contain these healthful or synergistic effects. The growing popularity of cannabis-derived products, especially the increasing popular vapes, are now bringing different groups and sources of terpenes into the cannabis world. In the world of cannabis products, terpenes within the product can be either cannabis-derived, other botanically derived, or artificial (synthetic). Products in which flavor and fragrance are heavily dependent, such as in vapes, concentrates and oils, some manufacturers will supplement or add other sources of terpenes to create a characteristic smell or taste especially when the extraction processes for some of these products can strip away many naturally-cannabis-derived compounds (7). As this process becomes more common for cannabis products the questions arise if the synthetic terpenes will have similar effects as the naturally-derived terpenes, and if the

Figure 2: Isopentenyl pyrophosphate.
Sample Processing and Analysis for Terpenes

As it was discussed in the last issue’s column regarding sampling and sample processing, cannabis products have a multitude of volatile compounds which must be retained during sample processing similar to the processing of economically valuable spices. Spices are another product rich in aromatic compounds such as terpenes. Spices and cannabis can be degraded by high temperatures and oxidation. In ambient temperature spice grinding processes, heat and energy are generated which can raise the temperature of spices to almost 100 °C and cause loss of critical aromatic components up to 60% loss (9,10). Reduction of temperature during processing can prohibit the breakdown of volatile compounds. In one study it was found that cryogenic conditions showed better retention of monoterpenes (myrcene, limonene, and pinene) than grinding at ambient temperature. These monoterpenes are the same primary monoterpenes in many cannabis varieties (11,12).

Table III: Terpene and terpenoid groups found in cannabis

<table>
<thead>
<tr>
<th>Terpene Group</th>
<th>Subgroup</th>
<th>MW</th>
<th>BP</th>
<th>Formula</th>
<th>Aroma</th>
<th>Reported Health Effects</th>
<th>Cannabis Variant Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Myrcenes</td>
<td>Monoterpene</td>
<td>136.2</td>
<td>168 °C</td>
<td>C_{10}H_{16}</td>
<td>Clove-like, musky earthy</td>
<td>Anti-inflammatory</td>
<td>Levorin 110, Skunk XL</td>
</tr>
<tr>
<td>Caryophyllenes</td>
<td>Sesquiterpene</td>
<td>204.4</td>
<td>264 °C</td>
<td>C_{15}H_{24}</td>
<td>Citrus</td>
<td>Anti-inflammatory; anti-convulsive; alcohol addiction</td>
<td>Gorilla Glue, Skywalker</td>
</tr>
<tr>
<td>Humulenes</td>
<td>Sesquiterpene</td>
<td>204.4</td>
<td>107 °C</td>
<td>C_{15}H_{24}</td>
<td>Hoppy</td>
<td>Anti-inflammatory; anti-cancer; insomnia; depression</td>
<td>Sour Diesel, Pink Kush</td>
</tr>
<tr>
<td>Pinenes</td>
<td>Monoterpene</td>
<td>136.2</td>
<td>156 °C</td>
<td>C_{10}H_{16}</td>
<td>Pine</td>
<td>Anti-inflammatory; anti-tumor; antioxidant; antidepressant</td>
<td>Bubba Kush, Strawberry Cough</td>
</tr>
<tr>
<td>Terpinolenes</td>
<td>Monoterpene</td>
<td>136.2</td>
<td>187 °C</td>
<td>C_{10}H_{16}</td>
<td>Pine with herbal and floral notes</td>
<td>Anti-inflammatory; anti-cancer; insomnia; depression</td>
<td>Pineapple Jack, Durban poison</td>
</tr>
<tr>
<td>Limonenes</td>
<td>Monoterpene</td>
<td>136.2</td>
<td>176 °C</td>
<td>C_{10}H_{16}</td>
<td>Lemon, citrus</td>
<td>Wound healing, antidepressant, anti-cancer</td>
<td>Lemon Haze, OG Kush</td>
</tr>
<tr>
<td>Carenes</td>
<td>Monoterpene</td>
<td>136.2</td>
<td>172 °C</td>
<td>C_{10}H_{16}</td>
<td>Pungent, earthy sweet</td>
<td>Anti-inflammatory</td>
<td>Super Silver Haze, Skunk #1</td>
</tr>
<tr>
<td>Phellandrenes</td>
<td>Monoterpene</td>
<td>136.2</td>
<td>172 °C</td>
<td>C_{10}H_{16}</td>
<td>Citrus &amp; Mint</td>
<td>Anti-inflammatory; antimicrobial</td>
<td>Super Lemon Haze, Super Silver Haze</td>
</tr>
<tr>
<td>Linalool</td>
<td>Monoterpene Alcohol</td>
<td>154.2</td>
<td>199 °C</td>
<td>C_{10}H_{15}O</td>
<td>Lavender &amp; Floral</td>
<td>Anti-inflammatory; antidepressant</td>
<td>Sour OG</td>
</tr>
<tr>
<td>Camphenes</td>
<td>Monoterpene Alcohol</td>
<td>136.2</td>
<td>159 °C</td>
<td>C_{10}H_{16}</td>
<td>Damp mint, pine notes</td>
<td>Anti-inflammatory; lowers cholesterol</td>
<td>Indica species</td>
</tr>
<tr>
<td>Terpineols</td>
<td>Monoterpene Alcohol</td>
<td>154.3</td>
<td>271 °C</td>
<td>C_{10}H_{15}O</td>
<td>Floral</td>
<td>Anti-cancer</td>
<td>Girl Scout Cookies, OG Crush</td>
</tr>
</tbody>
</table>
where many isomers or similar compounds are present. The most common method of analysis for terpenes is gas chromatography (GC) with either a flame ionization detector (FID) or mass spectrometer (MS).

The principles of gas chromatography is that samples are vaporized in an inlet at high temperatures of usually over 250 °C and transported via a carrier gas to a chemically infused column. The column material is composed of various chemical binding groups with interact with the vaporized analytes forcing the analytes out of the column phase over time and increasing temperature until the analytes are released into the carrier gas then the detector. The result is a chromatogram which displays graphical responses over time when each analyte is detected. Many GCMS column chemistries are based on boiling point or molecular size meaning that molecules of similar weight or similar boiling points could co-elute or appear overlapped in the chromatogram. It is this fact which can make the analysis of terpenes challenging since many terpenes have the same molecular weight and formula and relatively low boiling points which mean they appear very quickly in the chromatogram and often co-elute (Table III). Instrument and column manufacturers over the decades have become the experts on the separation of compounds and have given analytical labs many methods and specialized columns to aid in analysis.

Terpene analysis has always been an important component of many research areas including atmospheric chemistry, agricultural science, biochemistry, and environmental science to name a few. In industries dependent upon flavor and fragrance products, terpene profiles are part of their routine analytical testing procedures. The cannabis industry is just the newest industry to investigate the role of terpenes for flavor, fragrance and health benefits. As the industry continues to engineer cannabis strains to enhance specific chemical profiles, the importance of terpenes will increase.

References
11) S.M. Mathew, and V.V. Sreenarayanan, J. Spices Aromatic Crops 16(2), 82-87 (2007).
In this month’s column, we hear from three current and former state regulators about their collaborative work in the industry, feedback from stakeholders, and how they are making the industry more productive and safer for both businesses and consumers.

Joshua Crossney

Regulation Nation

Regulators play a vital role in an industry that is continuing to evolve.

I recently sat down with current and former state cannabis testing regulators to discuss challenges, changes and emerging efforts.

Featured here are Lori Dodson, who currently serves as the deputy director for the Maryland Medical Cannabis Commission; Deborah Miran, who was a member of the Natalie M. LaPrade Maryland Medical Cannabis Commission from 2013–2016; and Heather Krug, the state marijuana laboratory sciences program manager for the Laboratory Services Division of the Colorado Department of Public Health and Environment.

These industry leaders share their unique perspectives, insights and challenges, as well as comments on how cannabis laboratories are changing their practices.

What are some of the challenges that regulators face when crafting state regulations, given the federal status of cannabis and the fact that it is a states’ issue and regulations can vary greatly from state to state?

Lori Dodson: The obvious answer here is the lack of federal oversight leaves states essentially on their own deciding what is “safe” without cannabis data. States are left to review federal standards from United States Pharmacopoeia (USP) about botanicals, or tobacco, when trying to decide what level of contamination is considered an appropriate level. The American Herbal Pharmacopoeia (AHP) monograph was relied on pretty heavily, but the latest revision of that monograph was 2014. The pace of the industry makes it very difficult for the science to keep up. Additionally, the length of time required to promulgate regulations keeps states chasing the industry. I also believe there is such diversity in cannabis testing regulation because states are inadequately resourced at the staff level to scrutinize why certain regulations exist and whether or not they make sense scientifically. Many regulations are carbon copies of other states, regardless of adequacy.

Deborah Miran: The biggest challenge is a lack of a national standard for tests, methods, and specifications. Therefore, states are forced to rely on standards developed for other industries like traditional pharmaceutical testing, and medicinal herbs other than cannabis. The USP, even though they are a private, for-profit organization, is so closely aligned with the U.S. Food and Drug Administration (FDA) that they are blocked from publishing a monograph until cannabis is rescheduled.

Heather Krug: Regulators must work carefully when defining cannabis testing requirements to achieve a balance between protection of public health and safety and making testing too expensive, potentially driving black market activities. Regulatory testing requirements are best constructed upon evidence-based science, which can be difficult given the lack of currently available data specifically applicable to cannabis. This has resulted in variable regulations across states as each state has to define their own regulatory structure, with different state agencies playing various roles in that process. There have been many regulatory changes during the past few years of legalized marijuana, and the system will certainly continue to change as knowledge advances, hopefully in a way that begins to align across state lines.

What are some of the advancements that you have seen over the last three years in terms of expansion of products, qualifying conditions and routes of administration?

Dodson: I think the innovation with the processed product market is remarkable. There is a route of administration for everyone. Folks I talk to who are not familiar with medical cannabis still think the program is all about smoking a joint. I challenge them to really take a look at the innovation; it really mimics products available in pharmacies. The challenge with the rapid innovation is the ability for laboratories to keep up with the validation of methods to support all types of new products being developed. Sample preparation is drastically different for the variety of products available, and that equipment can be pricey for laboratories.

Miran: States seem to be very accepting of cannabis in traditional pharmaceutical dosage forms, and accordingly patients have access to medicine in a variety of routes of administration that they have familiarity with. While dried flower is still popular as a finished product, the processed products continue...
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States seem to be approaching qualifying conditions in widely varying ways. States like Maryland took a unique approach by listing broad categories of conditions rather than specific diseases, and this gives the medical discretion to the provider and patient rather than a state mandate. Accepting cannabis use for mental health conditions such as anxiety and depression has been difficult for regulators, and thus only a few states list anxiety and none list depression. Interestingly, in states that give wider discretion to the physicians, insomnia, depression, and anxiety are widely recommended.

**Krug:** Colorado has intended-use categories that allow for production of a broad array of product types. These categories include various inhaled products, orally consumed products, and skin and body products. It seems cannabis can be put into anything. Recently, we’ve adopted rules outlining requirements for production of certain higher-risk profile products including metered dose nasal sprays, inhalers and suppositories. We also have a process that allows producers to receive an alternative-use designation for products that don’t fit into any of the prescribed intended-use categories.

**Have you seen any changes in the cannabis testing industry based upon these advances?**

**Dodson:** The testing laboratories in Maryland have all been participating in laboratory comparison testing to ensure that all laboratories are able to prep, extract, and analyze all product types available. I hope to have a stability testing program available this year that will help define expiration dating for all product types. This will be especially important with the introduction of edibles in Maryland’s medical cannabis market.

**What is the biggest challenge that regulators are working to overcome at this moment?**

**Dodson:** Bridging the gap between the rule-makers, the laboratories and the industry. I’d love for industry players to begin to use the testing laboratories as resources to troubleshoot when a certificate of analysis doesn’t reflect the numbers that they believe it should, and not just immediately assume the laboratory is wrong. We are slowly getting there.

**Miran:** Sadly, as hard as regulators try and honest, legitimate laboratories try, there is still an unacceptable level of bad laboratory practices in many states. Often product pricing and licensee use, and cannabis in food products), accurate and precise testing of the active ingredients (cannabinoids and terpenes) becomes more challenging. To adequately validate a testing method, the producer of the dosage form must supply a placebo product (every ingredient minus the actives) to the testing laboratory in order to complete full method validation. In my experience this is rarely, if ever, happening, and this needs to change.

**Krug:** There are so many different matrices for laboratories to test because of the huge variety of product types. To address this challenge, we continuously see laboratories implementing new analytical, sample preparation and clean-up techniques specific to certain products, ranging from cryo-grinding, to filtration, to matrix-matched calibration. In response to advancements in the types of products being produced, Colorado also recently established new testing requirements specific to cannabis inhalers, nasal sprays, and suppositories to address potentially hazardous containments specific to these modes of administration.
profits are based on tetrahydrocannabinol (THC) content, so the incentive to "inflate THC results" in finished product testing exists.

Krug: From my perspective, one of the biggest challenges is trying to determine how to best protect patient and consumer health and safety in the absence of much of the data or the processes that typically exist for a non-cannabinoid product, such as established maximum residue levels for pesticides or approval of a new excipient. And just trying to keep up with the advancements of such an innovative and creative industry.

For newer states that are coming on-board and creating new state medical cannabis programs, what are some of the lessons learned when looking back at creation of particular commissions? What would you have done differently now?

Dodson: Adequate staffing is a must. Many different types of backgrounds are necessary to successfully regulate this industry. Our staff has grown from 17 personnel in December, 2017, to almost 50 today, and their backgrounds range from policy development and legal development to law enforcement, scientists, data analytics, IT, regulatory, and communications, just to name a few. To successfully regulate this industry there must be diversity in the regulatory staff. A second lesson learned is the necessity for thorough staff training. Think about it: cannabis regulators don’t come from a “pool of cannabis regulator candidates.” This industry is new and regulators across the country are still learning how to regulate appropriately. There is an inadequate amount of training for cannabis regulators that isn’t funded by the industry. I see this as an area with great potential if we are serious about doing our jobs well.

Miran: As exemplified in Maryland, I believe that a coordinated effort between a commission staff that reports to their Department of Health or pharmacy and an independent advisory board of experts is optimal. There are plenty of excellent examples of regulations in the 33 states that now have medical programs that no one needs to reinvent the wheel. Looking back, I would have favored a smaller, committed group of experienced experts to guide the staff decisions on medical and technical matters.

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Additionally, what are some of the successes that you participated in, and for which you are personally proud, that you would recommend being implemented in other states?

Dodson: I wrote the “Technical Authority for Medical Cannabis Testing” guidance document and hope it has been helpful for Maryland’s laboratories as well as other state testing programs that are coming on board. This document was written to provide a supplement to the regulations, and is meant to be revised as testing standards develop. We’ve also implemented a successful laboratory comparison testing program in Maryland. This program allows registered laboratories to compare their methods and analytics with other laboratories in the state (everyone is blinded, of course) on a variety of matrices. I see this program as a supplement to the proficiency testing requirement from ISO 17025. The sample preparation component can be very tricky when dealing with cannabis testing, so it’s critical that laboratories are able to use the appropriate matrix in addition to proficiency testing samples supplied by proficiency testing (PT) vendors when comparing testing competency.

Miran: Remember, I began regulating in 2013 and much has changed and improved since that time. Having said that, we accomplished many “firsts” in Maryland such as 1) requiring independent sampling of the product for testing, 2) requiring quality assurance (QA) retain and stability samples to be collected and tested as needed, and 3) adopting the FDA approach to the use of guidance documents for the details of testing rather than in regulations which are slow to revise when science advances.

Krug: We have made very deliberate efforts to engage our stakeholders in regulation and policy development. This has ranged from rulemaking, to designing and executing a multi-laboratory study on pesticide detection limits in cannabis, to establishing various work groups to focus on finding solutions to various issues. In August of 2018, we created the Marijuana Science and Policy Work Group whose purpose is to share ideas and information in a collaborative environment regarding the many scientific questions of relevance to the cannabis industry, to public health, and to public safety. Through public and private collaboration and study, it is hoped that the work group’s research, resources, and recommendations will be of value to industry, to regulators, and to policymakers even outside of Colorado. I’m proud to have been a part of all of these efforts. Not only do I think it’s allowed Colorado to be a leader in cannabis regulation, but it will allow us to help shape the cannabis industry of the future.

About the Interviewees

Deborah Miran, a former state commissioner, was a member of the Natalie M. LaPrade Maryland Medical Cannabis Commission from 2013-2016. While serving on the commission, she was also a member of the executive committee, policy, and research subcommittees, and was chair of the education subcommittee. She was responsible for developing education programs for doctors and patients, and was also an integral part of crafting the current regulations. Prior to the commission, she was president and founder of Miran Consulting, Inc.

About the Columnist

Joshua Crossney is the columnist and editor of “Cannabis Crossroads” and a contributing editor to Cannabis Science and Technology magazine. Crossney is also the president and CEO of CSC Events. Direct correspondence to: josh@jcanna.com
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Rob McCorkle, a retired lieutenant commander from a west coast law enforcement organization, was on a mission. His mother was diagnosed with brain cancer, and he was in search of an alternative to opioids for pain management. Having heard about the therapeutic benefits of medical cannabis, Rob connected with a couple of friends in the Portland area, which led to an investment to help them expand their medical cannabis grow, including funding its expansion up to a 10,000-square-foot recreational operation.

Around that same time, he re-connected with Chris Rushing, an old friend from the Department of Defense. Rushing had been working with hyper-spectral technology to track bomb makers in Iraq and Afghanistan, using special cameras to identify certain chemical signatures. Shortly afterward, Rushing introduced McCorkle to another colleague, Brendan Joyce. The two had been collaborating on a $40 million spectral imaging program for a large corn, wheat and soybean co-op in the Midwest, affixing multi-spectral cameras to the bottom of small planes to help farmers evaluate everything from water deficiencies to GMO corn infestations, and more.

“Given our synergistic backgrounds and collective expertise, we were able to figure out a way to apply the technology to build a spectral imaging service for the cannabis industry,” McCorkle says. “And Emerald Metrics was born.”

That was three years ago. Now with 10+ employees and a fully operational, proprietary CannaIntelligence™ software system, and Clonalyzer™ and CannabisCam™ hardware platforms for cannabis spectral imaging, Emerald Metrics is installing their systems in both medical and recreational grow operations in the US and Canada.

So, how does spectral imaging work?
“Every biologic in the world has a spectral code,” McCorkle says. “It’s like a fingerprint. Everything — people, plants, mold — all have a unique signature. So, we designed special cameras for plants, which can take a picture of a specimen with powdery mildew on it that is then identified in an imaging program using algorithms, and our proprietary artificial intelligence.”

One of the biggest benefits Emerald Metrics offers growers is assurance of plant health, generally in a range from worst (zero) to best (100). Using a multi-spectral clone camera, Emerald Metrics Clonalyzer™ takes real-time images of clones to identify which are the best to move forward for propagation.

“In my grow, I only keep clones that are 90 percent healthy and above,” McCorkle says. “The rest, I throw into the garbage. Last year, just using the Clonalyzer™ alone increased my yield by 12.7 percent.”

McCorkle believes the industry has done a great job looking at everything that goes into and around the plants. “But what no one in the industry is doing, except us, is looking at the actual plant itself — what the things you are doing are actually doing to the plant. We can tell you if you are harming or helping your plants so you can spend your resources wisely to ensure the best result.”

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Analysis of Veterinary Hemp-Based Oils for Product Integrity by LC/MS

Pet owners continue to seek alternative therapies for pain relief in their favorite companion animals. Instead of routine nonsteroidal anti-inflammatory drug (NSAID) treatment, there is a trend toward trying hemp-based products rich in cannabinoids. There are increasing numbers of companies producing hemp-based oils enriched with various mixtures of cannabinoids for pain and other maladies including seizures, cancer and anxiety. These products post labels attesting to the composition in the product, usually labeling for cannabidiol (CBD) concentrations in their products. But in the absence of regulatory control we questioned how accurate the information is on the label. The chemical composition of 13 commercially available oils intended for veterinary or a crossover of human and veterinary use was determined by selected ion monitoring liquid chromatography/mass spectrometry (SIM LC/MS). It was found that many of the labels were inaccurate regarding the cannabidiol concentrations and/or the presence of other cannabinoids. In general, the labels on most of the medicinal veterinary samples indicated higher levels of CBD than found in these studies by 20% or more. The precision and accuracy of SIM LC/MS analysis of the samples fell within the acceptable limits of regulated bioanalysis guidelines of +/- 15%.

Ben Nie, Jack Henion, and Joe Wakshlag

Many pet owners consider their animals beloved members of the family. When their ‘companion’ is not behaving normally or suffering from pain, they will seek professional help from their favorite veterinarian, and if that is not successful they will sometimes take it upon themselves to treat their ‘best friend’ with other remedies. Although tempting anecdotal reports on the benefits of cannabis- and hemp-based products continue to appear, most veterinarians remain reluctant to recommend cannabis to their clients for several reasons (1). Unfortunately, without proper research, the effects of medications containing CBD on animals remain unknown (2). Foremost is the fact that marijuana and hemp are still considered controlled substances by the U. S. Drug Enforcement Administration (DEA). The American Veterinary
Medical Association considers them illegal depending on the state laws, and discourages veterinarians from prescribing them for pets.

Although not currently considered pharmaceutical agents per se, there is clearly a lot of interest in the role that marijuana and hemp and their derived products may have in veterinary healthcare. Increasingly pet owners have already experimented with advertised medicinal cannabis products. These include oils reportedly containing therapeutic levels of CBD, in addition to other ingredients with product claims of beneficial effects. In states where recreational cannabis is legal, veterinarians are seeing increasing incidences of toxic cannabis cases in dogs. These popular companion animals have indiscriminate eating habits and often eat discarded cannabis products. In addition, veterinarians have had to respond to the pet owners’ queries regarding the proper use of medicinal cannabis. Since there is very little research to-date on these topics, veterinarians are often limited on their ability to wisely respond to these queries.

Medicinal marijuana, or medicinal cannabis, is a treatment regiment that has attracted considerable national attention recently (3). Controversy continues surrounding the legal, ethical, and societal implications associated with its use. Issues include safe administration, adverse health consequences and reported deaths attributed to marijuana intoxication. Therapeutic indications are based on limited clinical data and represent some of the complexities associated with this treatment. Cannabis indica or Cannabis sativa are considered marijuana or hemp. Regardless of what it is called, cannabis is currently considered by the DEA Comprehensive Drug Abuse Prevention and Control Act (Controlled Substances Act) of 1970 as a Schedule I controlled substance. This definition suggests cannabis has a high potential for abuse, no currently accepted medicinal use in treatment in the United States, and a lack of accepted safety data for use of the treatment under medical supervision (4). There is a dearth of research in support of its safety or therapeutic value in clinical settings.

Marijuana and hemp have been used medicinally worldwide for thousands of years (5). In the early 1990s, the discovery of cannabinoid receptors in the central and peripheral nervous systems created interest in other potential therapeutic values of marijuana (6). Since then, marijuana has been used by patients experiencing chemotherapy-induced anorexia, nausea and vomiting, pain, and forms of spasticity. Use among patients with glaucoma and human immunodeficiency virus (HIV) and acquired immune deficiency syndrome (AIDS) has also been widely reported (7). It should not be surprising that veterinarians may be interested in the potential for medicinal cannabis to relieve pain and other clinical issues associated with their animal patients (8,9).

The historically restrictive legal guidelines for cannabis have significantly limited research on its safety and efficacy in both humans and animals (10). However, a recent study by researchers at the Cornell University College of Veterinary Medicine reported on the potential beneficial effects of CBD oils administered orally to osteoarthritic (OA) dogs (11). A representative commercial veterinary medicinal CBD oil was purchased and administered orally in the food of known OA canine patients. The publication described the pharmacokinetics, safety and clinical efficacy of cannabidiol treatment in osteoarthritic dogs, which suggested that a 2 mg/kg of CBD twice daily can help increase comfort and activity in dogs with this condition. These findings spurred us to examine whether the selected 13 commercial oils were similar in chemical composition. In this report we describe the chemical analysis of some representative commercial veterinary oils by SIM LC/MS to determine the concentration of the constituents reported on the labels of those products.

This analytical approach is one of several available for such an application as described in a recent review (12). We also monitored common other cannabinoid constituents which were not listed on some of the product labels. There are an ever-increasing number of suppliers of such oils and related products which are sold in the absence of any regulatory control.

The goal of this study was to compare our analysis results on the cannabinoid composition with the indicated concentrations of selected compounds on the product label. A recent report on phytocannabinoids common to the cannabis cultivars present in medicinal oils for human use was reported, where high performance liquid chromatography/ultra violet detection (HPLC/UV) analyses were described (13). The report described herein expands upon hemp oil analysis with a focus on commercially available oils intended for pet consumption using the more sensitive and selective analytical capabilities of SIM LC/MS.

Experimental

Instrumental and Chromatographic Conditions

The mass spectrometer employed in this report was the Expression L-model single quadrupole system (Advison, Ithaca, NY). The mass spectrometer was operated in the SIM mode using electrospray ionization coupled with LC/MS while monitoring the characteristic ions of cannabidiolic acid (CBDA) (m/z 341.3), cannabinerol (CBG) (m/z 317.2), CBD (m/z 315.3), cannabinol (CBN) (m/z 313.3), THC (m/z 315.3) and tetrahydrocannabinolic acid (THCA) (m/z 341.3). The positive ion mode was employed with a capillary temperature...
of 250 °C, capillary voltage of 180 V, a source span of 0, a source offset of 25 V and a source gas temperature of 250 °C. The chromatographic conditions used employed an Avant binary HPLC system (Advion, Ithaca, NY) equipped with a Restek Raptor ARC 18 (3.0 mm x 100 mm) column packed with 1.8 micron particles. The binary gradient mobile phase consisted of 0.1% formic acid in water (mobile phase A) and 0.1% formic acid in acetonitrile (mobile phase B). The initial HPLC conditions commenced with 75% B programmed to 95% B over two min which was then held isocratic at 95% B for a total run time of 3.5 min. After this gradient program the mobile phase was recycled over 2 min to the initial 75% B conditions. The injection volume was 20 microliters, the flow rate was 0.6 mL/min and the column temperature was maintained at 30 °C.

**Hemp Oil Sample Preparation**

Ten μL of each oil sample were added to 490 μL of isopropyl alcohol and mixed on a vortex mixer to dissolve into a uniform solution for each oil sample. Fifty μL of this initial diluted oil sample in isopropyl alcohol were then further diluted into 450 μL of 50% aqueous methanol. These secondary dilutions were then filtered through 0.2 μm syringe filter into the respective HPLC vials for SIM LC/MS analysis (total dilution factor x 100 mm) column packed with 1.8 micron particles. The binary gradient mobile phase consisted of 0.1% formic acid in water (mobile phase A) and 0.1% formic acid in acetonitrile (mobile phase B). The initial HPLC conditions commenced with 75% B programmed to 95% B over two min which was then held isocratic at 95% B for a total run time of 3.5 min. After this gradient program the mobile phase was recycled over 2 min to the initial 75% B conditions. The injection volume was 20 microliters, the flow rate was 0.6 mL/min and the column temperature was maintained at 30 °C.

**Standard Curves**

One milliliter of coconut oil was dissolved in 49 mL of isopropyl alcohol. The coconut oil was used as a representative negative control matrix relative to the veterinary oils. Five mL of this isopropyl solution of coconut oil was further diluted with 45 mL of 50% aqueous methanol. This diluted coconut oil was used as the solvent matrix for preparation of calibration curves for the six targeted cannabinoids. The latter contained CBDA, CBG, CBD, CBN, THC and THCA which produced a calibration curve with concentrations of 1, 5, 10, 50, 100, 500, 1000, 5000, 10,000, and 20,000 ng/mL for each cannabinoid compound. Ten μL of the internal standards (CBD-d3, CBN-d3 and THC-d3 at 1000 ng/mL) were added into 200 μL of calibrators containing each of the six targeted cannabinoids. The internal standard final concentrations were 47.6 ng/mL. Quality control (QC) samples were prepared in the same manner independently at concentrations of 10, 100, 1000, and 10,000 ng/mL fortified with the same stable isotope internal standards as calibrators. The QC samples were analyzed by SIM LC/MS in six replicates within the day of analysis to produce the intra-day precision and accuracy values shown in Table I. It is noted in Table I that the accuracy for the targeted cannabinoids are in general quite acceptable pursuant to regulated bioanalysis guidelines, with the exception of THCA. These results are in agreement with another report which employed LC/MS/MS techniques (14). This may result from the absence of a stable isotope internal standard for this compound or it may suggest that negative ion detection may be a better choice for this application. Additional experimentation may be required to better understand this unexpected discrepancy.

**Results and Discussion**

Our objective in this work was to analyze a representative set of commercial veterinary CBD oils to determine whether the labeled contents were accurate. Also, in the absence of any regulatory control, we were interested to learn whether the THC content in these samples was within legal limits of, for example, the Canadian 10 parts per million (ppm) guidelines.

There is a long list of the naturally occurring chemicals occurring in cannabis plants (15). Cannabis is very complex in its chemistry due to the vast number of its constituents and their possible interaction with one another. These compounds represent many of the chemical classes, for example, mono- and sesquiterpenes, sugars, hydrocarbons, steroids, flavonoids, nitrogenous compounds and amino acids, among others. However, hemp oil extracts usually contain relatively high (several percent) concentrations of CBD and the major carboxylic acids along with much lower levels of other cannabinoids along with a wide range of many other chemicals (16).
The final composition of commercial hemp oil will depend upon the type of extraction used and any subsequent sample extract purification or treatment. The relative amounts of these chemicals will also depend upon the hemp cultivar selected (6).

Figure 1 shows the chemical structures for six common cannabinoids present in hemp oil that were measured in this study (rows 1 and 3). Also shown in Figure 1 are the structures for three of the stable isotope incorporated internal standards used for quantitative analysis (row 2). These include THC-d3, CBD-d3, and CBN-d3.

These three internal standards were added to the diluted hemp oil samples at concentrations in the lower quartile of the calibration curve.

The LC/MS analysis of these samples produced LC/MS chromatograms with these stable isotope labelled internal standards co-eluting with their non-deuterated analogs while displaying the same chemical behavior through the chemical analysis procedure.

They may be easily differentiated from their non-labelled analogs via their mass differences which are 3Da higher in each case.

Shown in Table II is a summary of the concentrations determined by SIM LC/MS of the six targeted cannabinoids shown in Figure 1 measured in veterinary CBD oils of this study which are labelled Vet CBD Oil samples 1-13. As a representative example, the chromatogram shown in Figure 2 was obtained from the SIM LC/MS bioanalysis of medicinal veterinary oil sample number 8 (Table II). Employing the experimental conditions described (vide supra) the analysis of each sample was completed within three min and clearly shows the six targeted cannabinoids in addition to a few additional chemical constituents in the oil sample. The simplicity of this chromatogram is due in part to the additional selectivity provided by a mass spectrometer detector operated in the SIM LC/MS acquisition mode. It is interesting to note in Figure 2 the relatively high level of THC in this sample (when...
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- Ethan Russo, MD (Director, R&D, ICCI)
there was no reference on the product label that THC was present in the sample observed at a retention time of 2.33 min in contrast to the expected high concentration of CBD shown with its retention time of 1.68 min. With reference to Table II the corresponding concentrations for these compounds were 0.348 mg/mL for THC and 15.5 mg/mL for CBD, respectively.

The cannabinoid concentrations observed in the medicinal veterinary oil samples ranged from not detectable (N/D) to very low concentrations for CBDA, CBN and CBG. This perhaps is not too surprising as it is generally known that some of these cannabinoids are present at relatively low levels in cannabis and hemp (7). An exception is sample number 12 which had a concentration of 11.8 mg/mL for CBDA. This sample also had elevated levels of some other cannabinoids. It is unclear why sample number 12 would have such elevated levels for some of the cannabinoids based upon the available information, but it still falls under the 2018 Farm Bill’s limits of less than 0.3% THC as a hemp product.

Since these medicinal veterinary oils were marketed as CBD oils, it was anticipated that the CBD content would be elevated in the samples. CBD has received considerable “press” as a potentially useful drug for a variety of ailments (4). Recently the U.S. Food and Drug Administration (FDA) approved Epidiolex, which is CBD derived from its extraction of the marijuana plant (17). The CBD column in Table II shows the CBD concentrations across the 13 samples ranging from 0.88 mg/mL for sample number 6 up to 27.5 mg/mL for sample number 12.

Looking at the concentration of CBD in the rest of 13 medicinal veterinary oils listed in Table II it is interesting to note the range of concentrations observed. To help see this more graphically one may look at the bar graph shown in Figure 3. This figure shows a comparison of the CBD concentrations in all 13 medicinal oil samples (Table II) determined by SIM LC/MS compared with the indicated levels of CBD on the product label. The black bars in Figure 3 graphically shows the vendor-reported concentration in mg/mL (the concentration units often shown on the product labels) for CBD in each of the 13 medicinal oil samples. Adjacent to each black bar is a solid grey bar showing the SIM LC/MS experimentally derived concentration for CBD in each sample. In samples numbered 3, 4, 7, 9, 10, and 12, the concentrations for CBD were reported to be substantially higher (black bars) on the label than the actual measured concentration measured by SIM LC/MS in this study (grey bars). Thus, the experimental results show the amount of CBD in the oil is often lower (grey bars in Figure 3) than the amounts reported on the label (black bars in Figure 3) except for

<table>
<thead>
<tr>
<th>Sample Number</th>
<th>CBD label</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>25 mg hemp extract/ml</td>
</tr>
<tr>
<td>2</td>
<td>N/A</td>
</tr>
<tr>
<td>3</td>
<td>200 mg/oz</td>
</tr>
<tr>
<td>4</td>
<td>10 mg/mL</td>
</tr>
<tr>
<td>5</td>
<td>4.2 mg/mL</td>
</tr>
<tr>
<td>6</td>
<td>56 mg hemp leaf extract/ml</td>
</tr>
<tr>
<td>7</td>
<td>250 mg cannabinoids</td>
</tr>
<tr>
<td>8</td>
<td>5 mg of hemp oil/0.5 ml</td>
</tr>
<tr>
<td>9</td>
<td>350 mg/30mL</td>
</tr>
<tr>
<td>10</td>
<td>225 mg total in 15 ml</td>
</tr>
<tr>
<td>11</td>
<td>150 mg/18 ml</td>
</tr>
<tr>
<td>12</td>
<td>website 3000 mg/60 ml</td>
</tr>
<tr>
<td>13</td>
<td>8 mg/mL</td>
</tr>
</tbody>
</table>

**Table II:** Summary of veterinary CBD oil concentrations for the six cannabinoids measured in samples 1-13. Concentrations are mg/mL of indicated cannabinoid. It should be noted that sample 6 was a crude hemp leaf extract without subsequent sample cleanup or concentration.

<table>
<thead>
<tr>
<th>Vet CBD Oil</th>
<th>CBDA</th>
<th>CBG</th>
<th>CBD</th>
<th>CBN</th>
<th>THC</th>
<th>THCA</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.005</td>
<td>0.070</td>
<td>13.4</td>
<td>0.078</td>
<td>0.347</td>
<td>0.107</td>
</tr>
<tr>
<td>2</td>
<td>0.007</td>
<td>0.051</td>
<td>10.6</td>
<td>0.005</td>
<td>0.194</td>
<td>0.026</td>
</tr>
<tr>
<td>3</td>
<td>0.001</td>
<td>N/D</td>
<td>1.65</td>
<td>0.030</td>
<td>0.023</td>
<td>0.075</td>
</tr>
<tr>
<td>4</td>
<td>N/D</td>
<td>N/D</td>
<td>6.51</td>
<td>0.003</td>
<td>0.001</td>
<td>0.088</td>
</tr>
<tr>
<td>5</td>
<td>0.042</td>
<td>0.026</td>
<td>4.34</td>
<td>0.018</td>
<td>0.103</td>
<td>0.076</td>
</tr>
<tr>
<td>6</td>
<td>0.153</td>
<td>N/D</td>
<td>0.88</td>
<td>0.002</td>
<td>0.014</td>
<td>0.244</td>
</tr>
<tr>
<td>7</td>
<td>1.42</td>
<td>0.061</td>
<td>5.76</td>
<td>N/D</td>
<td>0.205</td>
<td>0.049</td>
</tr>
<tr>
<td>8</td>
<td>0.291</td>
<td>0.274</td>
<td>15.5</td>
<td>0.062</td>
<td>0.348</td>
<td>0.180</td>
</tr>
<tr>
<td>9</td>
<td>0.087</td>
<td>0.147</td>
<td>8.82</td>
<td>0.009</td>
<td>0.278</td>
<td>N/D</td>
</tr>
<tr>
<td>10</td>
<td>N/D</td>
<td>0.102</td>
<td>11.6</td>
<td>0.017</td>
<td>0.352</td>
<td>N/D</td>
</tr>
<tr>
<td>11</td>
<td>N/D</td>
<td>0.018</td>
<td>7.86</td>
<td>N/D</td>
<td>0.049</td>
<td>0.079</td>
</tr>
<tr>
<td>12</td>
<td>11.8</td>
<td>0.372</td>
<td>27.5</td>
<td>0.005</td>
<td>1.29</td>
<td>0.693</td>
</tr>
<tr>
<td>13</td>
<td>0.123</td>
<td>0.116</td>
<td>13.9</td>
<td>N/D</td>
<td>0.374</td>
<td>0.032</td>
</tr>
</tbody>
</table>

**Table III:** A listing of the indicated concentrations on the product labels of the 13 veterinary medicinal oils studied. The lack of uniform guidelines or practices for labelling medicinal CBD oils leads to considerable confusion for customers.
oils 5 and 13. In general, the manufacturers for these products are labeling CBD concentrations higher than the content actually present in the product. However, it is encouraging to note that some vendors provide accurate indications of the CBD contents as noted for the measured CBD concentrations in samples 5, 9 and 11 which were close to the label indications. It is sobering to note that some vendors label their product contents such that the buyer is unable to know the total cannabinoid or individual cannabinoid concentrations. For example, for samples 1, 2, 6 and 8 the indicated concentration on the bottle was “mg of hemp extract per milliliter.” This claim provides the buyer no idea of the actual levels of cannabinoids in the product. Clearly a systematic format for indicating the concentration content would be helpful for the buyer. Table III shows the variety in the CBD label nomenclature used for the 13 samples used in this study.

Since THC is the psychoactive component in cannabis and hemp that has been a concern in the recent past, it is important to know that the concentration of THC in medicinal oils and products is below government action levels. In Canada, the level of THC in such samples is limited to 10 ppm (10 microgram/gram), while the U.S. 2018 Farm Bill suggests that THC concentrations of hemp products must be less than 0.3%. In the absence of any regulatory authority the customer has no way of knowing whether the vendor abides by the established guidelines. The experimentally determined THC concentrations listed in Table II are plotted in Figure 4 with reference to the units of ppm. A horizontal red dashed line is plotted at the 10 ppm THC level allowed by Canada in such samples. One can see that only sample 4 in this study contains THC below this 10 ppm level. Most of the samples have THC content well above the 100 ppm level with sample 12 weighing in close to 1300 ppm. It is worth noting the bottle label on sample number 12 as well as the other samples analyzed in this study made no reference to THC being in the product. These results suggest the need for dependable accuracy and consistency in the labelling of veterinary or possibly other medicinal CBD oils and products.

To help the reader gain a better understanding of the ‘analytical view’ of these results that is not so obvious in tabulated data we reference the results shown in Figure 5. This figure shows the

![Figure 2: SIM LC/MS total selected ion current chromatogram for veterinary CBD oil sample number 8. In the order of LC/MS elution time the quantities measured in mg/mL were: CBDA = 0.291; CBG = 0.274; CBD = 15.5; CBN = 0.062; THC = 0.348; and THCA = 0.180.](image)

**Figure 2:** SIM LC/MS total selected ion current chromatogram for veterinary CBD oil sample number 8. In the order of LC/MS elution time the quantities measured in mg/mL were: CBDA = 0.291; CBG = 0.274; CBD = 15.5; CBN = 0.062; THC = 0.348; and THCA = 0.180.

![CBD content](image)

**CBD content**

<table>
<thead>
<tr>
<th>On Label</th>
<th>Measured by LC/CMS</th>
<th>Exceed +/- 20%</th>
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</tbody>
</table>

![Figure 3: This figure shows a comparison of the CBD concentrations in all 13 medicinal oil samples determined by SIM LC/MS compared with the indicated levels on the product label. It was not possible to know the labeled CBD concentrations for Samples 1, 2, 6 and 8 due to the units used which were indicated to be mg of hemp extract/mL. The error bars depict the degree of accuracy in these measurements. The accuracy and precision is quite good due in part to the relatively high concentrations of the target analytes in the samples and the high sensitivity and selectivity of the SIM LC/MS analytical technique. The samples with asterisks denote that the labels were more than plus or minus 20% in error of the actual concentration of CBD in the sample.](image)

**Figure 3:** This figure shows a comparison of the CBD concentrations in all 13 medicinal oil samples determined by SIM LC/MS compared with the indicated levels on the product label. It was not possible to know the labeled CBD concentrations for Samples 1, 2, 6 and 8 due to the units used which were indicated to be mg of hemp extract/mL. The error bars depict the degree of accuracy in these measurements. The accuracy and precision is quite good due in part to the relatively high concentrations of the target analytes in the samples and the high sensitivity and selectivity of the SIM LC/MS analytical technique. The samples with asterisks denote that the labels were more than plus or minus 20% in error of the actual concentration of CBD in the sample.
SIM LC/MS chromatograms for two of the samples analyzed in this study. It is worth noting these chromatograms reveal relatively few peaks or chromatographic components due in part to the rather high concentrations of the targeted cannabinoids as well as the fact that SIM acquisition experiments measure only those m/z values selected for the experiment. Figure 5a shows the SIM LC/MS chromatogram for medicinal oil sample number 6 which as shown in Figure 4 had a THC concentration just above the 10 ppm allowed level. As is apparent in Figure 5a the THC peak at 2.34 min retention is a minor component along with the other minor cannabinoids with the obvious exception of the large CBD peak at a retention time of 1.68 min retention time. However, in Figure 5b the THC peak in sample 12 is a much more abundant component relative to the other cannabinoids with again the exception being the CBD peak at 1.68 min retention time. Note that the y-scale in both Figure 5a and Figure 5b is the same such that the reader can directly compare the relative quantities of THC in these two samples. The authors suggest the quantitative determination of cannabis constituents in medicinal products can be reliably acquired from results such as those shown in Figure 5.

This report has highlighted label discrepancies for the commercial veterinary medicinal CBD oil products. In contrast, Table IV shows concentration data for a different commercial medicinal cannabis sample where the vendor’s reported concentrations appear to be quite accurate. The vendor provides analytical concentration results for the product cannabinoids from two independent laboratories which agree quite closely. Accordingly, the SIM LC/MS procedure described in this report was also employed with this product and our results concur with those provided by the vendor. The first column in Table IV shows the six cannabinoids reported to be contained in the product. The second column summarizes the results obtained by the authors’ laboratory while columns 3 and 4 in Table IV are the analytical results from two other independent laboratories labelled Laboratory 1 and Laboratory 2. These three sets of bioanalysis results provide confidence to the customer that the product composition is accurate for this particular product.

Conclusions
The potential benefit of medical marijuana or hemp nutraceuticals for veterinary patients is attracting increased attention, but many veterinarians remain relatively uninformed and somewhat skeptical of recommending cannabis-derived products to their customers. Researchers and practitioners would welcome clinical research on the medical utility, safety and efficacy of marijuana-derived products. Instead, many companion animal owners are taking it upon themselves by responding to commercial claims that medicinal oils and products may help alleviate pain and other ailments of their favorite pet. This can lead to the purchase and administration of unproven or unreliable medications or even potentially dangerous treatments that may not help...
FDA-approved medicines provide patient, pesticide contamination or heavy metal limits.

Simple yet major questions for a consumer are: Does the product contain what is advertised; Is it safe?, and Is it effective? A lot of research is still needed to answer these questions, although it is entirely possible to provide accurate chemical analysis of the product composition as this publication has attempted to demonstrate. Just as the last example described in Table IV, a reputable company can provide accurate composition data if they are legitimate. The analytical technology described in this report is entirely capable of providing this information if the vendor has the integrity to employ it.

Acknowledgements

The authors thank their Advion colleagues, Drs. Daniel Eikel and Changtong Hao, for helpful discussion and contributions to this work as well as Mr. Seth Richardson of Kingsland Partners, Inc. for helpful input on hemp plant biology.

References

16) C. Giroud, Chemia, 56 (3), 80-83 (2002).
17) U.S. Food and Drug Administration (FDA) “FDA approves first drug comprised of an active ingredient derived from marijuana to treat rare, severe forms of epilepsy. https://www.fda.gov/NewsEvents/Newsroom/PressAnnouncements/ucm611046.htm
Beyond $\Delta^9$-THC and CBD: Current Evidence for Medical Benefit of Terpenes and Less Studied Cannabinoids

Cannabis sativa has been used medicinally for more than 4000 years. With resurgent interest in medical cannabis, 31 states have approved its use by patients with various conditions. Unlike conventional pharmaceuticals, cannabis varies widely in the abundance of medically beneficial compounds. Due to federal prohibition, little information is available to medical professionals, patients, or dispensary staff concerning specific components of cannabis and their known medicinal properties. This can result in patients choosing cannabis products with molecular profiles that are not well-suited to their precise medical needs. Besides the two main active compounds in C. sativa, 9-delta-tetrahydrocannabinol ($\Delta^9$-THC) and cannabidiol (CBD), other cannabinoids and aromatic molecules called terpenes have demonstrated therapeutic effects. Here we explore evidence that lesser-known cannabinoids and terpenes may be useful to patients with specific conditions. This information should facilitate better recommendations to patients and empower them to choose effective medical cannabis products.

Victoria Allen

Although Cannabis sativa contains more than 483 different phytochemicals (1), most medicinal properties of the plant are thought to result from specific cannabinoids and terpenes, acting both discretely and synergistically. The most well-known cannabinoids are 9-delta-tetrahydrocannabinol ($\Delta^9$-THC) and cannabidiol (CBD). Preparations of these compounds have been studied in randomized, placebo-controlled clinical trial with favorable outcomes. $\Delta^9$-THC has been approved by the U. S. Food and Drug Administration (FDA) for treating both anorexia associated with HIV and AIDS and nausea and vomiting because
of chemotherapy (2), while CBD has been approved by the FDA as an antiseizure medication (3,4). These two major cannabinoids have also shown clinical benefits in eight human trials for the reduction of chronic pain (neuropathic pain and cancer pain), in five human trials for the reduction of spasticity because of multiple sclerosis or paraplegia, and also in human trials for anxiety disorder, sleep disorder, psychosis, and Tourette’s syndrome (5). However, there is also evidence that other, comparatively lesser studied cannabinoids may have therapeutic potential.

All phytocannabinoids are aromatic, oxygen-containing hydrocarbons. They are derived from a cannabigerol (CBG)-like skeleton, consisting of a benzene ring bound to two oxygen atoms and a 3- or 5-carbon propyl- or pentyl-chain (Figure 1). The various cannabinoids display unique properties mainly because they differ in how the CBG-skeleton is cyclized. In addition to CBD and Δ2-THC, dozens of other naturally occurring cannabinoids have been identified in C. sativa (1). The cannabinoids are thought to have differing physiological effects based on their varied interactions with the known cannabinoid receptors CB1 and CB2, although phylogenetic evidence suggests that additional cell membrane-bound receptors may also respond to cannabinoids (6). Consistent with this theory, several receptor knockout experiments have shown cannabinoids can exert their effects even when treated cells are missing the CB1 or CB2 receptor (7–9).

Cannabis terpenes are organic molecules made up of one or more 5-carbon isoprene units. C. sativa produces mainly monoterpenes (two isoprene units) and sesquiterpenes (three isoprene units), which make up 48–92% and 5–49% of the plant’s terpene content, respectively (1). Terpenes, which are extremely volatile, are responsible for the odor and flavor of cannabis. C. sativa is only one of many plants that produce terpenes; in fact, terpenes are the primary active constituent in most essential oils (EOs). Because many terpenes are synthesized by plants other than C. sativa, comparatively more data on terpenes is available than on alternative cannabinoids. However, much of the available data is derived from studies on EOs and not pure terpenes (9). Although in some cases, a particular terpene makes up as much as 95% of the EO tested (10), we cannot exclude the possibility that another chemical constituent of the EO acting in synergy with that terpene is responsible for the pharmacological effects observed.

Both cannabinoid and terpene production depend on strain, sex, age, plant part, and growth conditions. Harvest methods and postharvest conditions also affect terpene and cannabinol (CBN) content (1). Terpenes and cannabinoids are both mainly stored in glandular trichomes (11,12). Terpenes constitute up to 4% of cannabis flower dry weight, while cannabinoids constitute up to 30% (13).

The following paragraphs will discuss health benefits of specific cannabinoids and terpenes. It is important to keep in mind cannabis science is an emerging field and most available data is extremely preliminary. Double-blind, randomized, placebo-controlled clinical trials have not been conducted, limiting our ability to draw conclusions at present. Almost all evidence

Figure 1: Structures and biosynthetic pathway of cannabinoids. (Adapted and reprinted with permission from J. Nat. Prod. 2016.)
collected to date on cannabinoids is either from animal models, cell culture (in vitro), or purely biochemical or molecular. It is therefore impossible to extrapolate even approximate human dosage equivalents from the most rigorous of these studies. This is especially true given animal studies most often administer drugs of interest intravenously, subcutaneously, or parenterally, whereas patients often administer medical cannabis products through inhalation. Data from in vitro studies is equally limited in its utility for human patient applications, often drawing conclusions from only one cell type that is incubated in a container for a specific length of time with specific concentrations of the compound of interest. Molecular and biochemical data (for example, receptor binding, gene, and protein expression) will not be considered in this review because of the even greater difficulty of providing context for their interpretation.

Compounding the challenges of interpreting in vitro and animal studies is the variability in cannabinoid and terpene content in different cannabis strains and preparations. Clinical trials involving multiple commercial strains, alternative medical cannabis products, and single and polycompound extracts are urgently needed. Crucially, the lack of human studies signifies an absence of data on adverse effects or events. Therefore, it is imperative that patients should only attempt to medicate with cannabis products while under the supervision of an experienced and knowledgeable physician.

**Cannabichromene**

Cannabichromene (CBC) showed anti-inflammatory properties in both a rat paw edema (swelling) test (14) and a murine colitis assay (15,16), which is consistent with anti-inflammatory effects observed in murine cell culture (15,16). CBC also inhibits cancer growth in several different cell lines as well as reducing lung metastases from cancer cell injections, but has a markedly weaker effect than CBD (17). CBC was shown to have a weak analgesic effect in a mouse model, but not as strongly as ∆²-THC (18). Most promisingly, microbial culture assays show CBC has strong antibacterial properties, even stronger than antibiotics in some cases, and rivaling that of ∆⁹-THC and CBD (19). This study also showed that CBC has mild antifungal activity. CBC strongly inhibited drug-resistant Staphylococcus aureus (MRSA). However, this effect was not unique to CBC (20). These antimicrobial studies used several different versions of synthesized CBC, and found that antimicrobial effects varied depending on the exact arrangement of the molecule (Figure 2). Therefore, extracted CBC preparations will likely have varying efficacies depending on the relative abundance of the different chemical analogues.

**Cannabigerol**

Cannabigerol (CBG) compounds were also found to exert strong antibacterial and mild antifungal properties in cell culture (19). CBG also strongly inhibited MRSA growth, although CBD had slightly higher efficacy (20). Similar to CBC, CBG inhibited cancer cell growth in several cell lines (17,21), but not to the extent that CBD did. Similarly, CBG inhibited keratinocyte proliferation, implying a possible use against psoriasis and acne (7). Although some reviews cite CBG studies that showed antidepressant or antihypertensive effects, these works were not peer-reviewed.

**Cannabinol**

Cannabinol (CBN) showed weak anticonvulsant activity compared to ∆⁹-THC and CBD in a murine model (22), although vehicle-treated animals were not included in the data shown (it is assumed here that electroshocks reliably induced convulsions in 100% of vehicle-treated animals). CBN inhibited pentobarbital metabolism in vitro, although not to the extent of CBD (23), however another study found little effect on pentobarbital metabolism (24). Curiously, a more recent study showed that CBN-treated mice slept about twice as long as controls after pentobarbital administration (25), indicating that CBN may actually enhance the sedative effects of other drugs.

CBN also had an analgesic effect in a mouse model, but it was weaker than that of low dose aspirin (26). CBN showed pronounced antibacterial activity against several MRSA strains (20). CBN also inhibits keratinocyte proliferation at dosages similar to CBG, ∆⁹-THC, and CBD (7), indicating potential therapeutic use in acne and psoriasis. CBN also showed substantial (almost three-fold) appetite-stimulating effects in a rat model (8). There is also evidence that CBN can act as a vasorelaxant in isolated rat hepatic arteries (27). Claims that CBN stimulates bone formation are not based on peer reviewed research; in fact, a peer-reviewed study found that the effects of CBN on bone marrow in live mice were “not clearly interpretable” (28).

**Cannabidivarin**

Cannabidivarin (CBDV) has some of the strongest evidence favoring a therapeutic effect. First studied in rodent models, it reduced seizure frequency by about 40% in rat hippocampal slices (in vitro), and had an even greater effect of reducing seizures in live mice (29). Notably, CBDV also reduced mortality in two models of seizure induction in both mice.
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and rats; in one rat model, mortality in CBDV-treated animals was reduced to less than 20% compared to controls (29). Although CBD and CBDV show comparable anticonvulsant activity in different animal studies (30), they have not been compared in a single study. It remains to be determined whether individual patients may experience greater efficacy or improved tolerance from CBDV than from CBD, but a pharmaceutical company is already patenting the use of CBDV for treatment of seizures (31). The same company has initiated Phase II clinical trials of a CBDV preparation (32) after Phase I trials revealed a “reassuring safety profile” with a lack of adverse events. CBDV has also shown an antinausea effect in a rat model, reducing the nausea-index behavior by about a third (33).

<table>
<thead>
<tr>
<th>Terpene</th>
<th>Associated Fragrance or EO</th>
<th>Selective Putative Medicinal Effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>α-Pinene</td>
<td>Conifers</td>
<td>Bronchodilator, Irritant</td>
</tr>
<tr>
<td>Camphene</td>
<td>Conifers, especially Douglas fir</td>
<td>AO</td>
</tr>
<tr>
<td>β-Pinene</td>
<td>Conifers</td>
<td>AO</td>
</tr>
<tr>
<td>Myrcene</td>
<td>Earthy, musky</td>
<td>AO, analgesic</td>
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<tr>
<td>α-Phellandrene</td>
<td>Frankincense</td>
<td>Analgesic</td>
</tr>
<tr>
<td>Carene</td>
<td>Turpentine, white pepper</td>
<td>Eye, nose, skin and lung irritant</td>
</tr>
<tr>
<td>α-Terpinene</td>
<td>Tea tree oil</td>
<td>AO, AP</td>
</tr>
<tr>
<td>Limonene</td>
<td>Citrus</td>
<td>AB, AO, antidepressant, anti-inflammatory, anxiolytic</td>
</tr>
<tr>
<td>β-Ocimene</td>
<td>None, social-regulatory pheromone of honeybees</td>
<td>Anticonvulsant*, antitumor*</td>
</tr>
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<td>Eucalyptus, cumin</td>
<td>AB, AO</td>
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<tr>
<td>Terpinolene</td>
<td>Conifers</td>
<td>AB, AO</td>
</tr>
<tr>
<td>Linalool</td>
<td>Lavender, basil</td>
<td>Anticonvulsant, antimicrobial?</td>
</tr>
<tr>
<td>Fenchol</td>
<td>Basil</td>
<td>None reported</td>
</tr>
<tr>
<td>α-Terpineol</td>
<td>Tea tree oil</td>
<td>AB, AP</td>
</tr>
<tr>
<td>β-Caryophyllene</td>
<td>Oregano, cloves, hops, rosemary, black pepper</td>
<td>AB, AF, AO</td>
</tr>
<tr>
<td>α-Humulene/α-Caryophyllene</td>
<td>Hops (Humulus lupus)</td>
<td>AO</td>
</tr>
<tr>
<td>Caryophyllene oxide</td>
<td>Lemon balm, eucalyptus</td>
<td>AB, AF</td>
</tr>
</tbody>
</table>

*AB = antibacterial, AF = antifungal, AO = antioxidant, AP = antiprotozoan. All reviewed in reference 9. * = Not yet studied in humans.

**Tetrahydrocannabinivarin**

Rock and colleagues (33) also demonstrated that Δ9-thetahydrocannabinivarin (Δ9-THCV) completely blocked the behavioral effects of nausea and reduced saccharin palatability, indicating a possible antiappetite effect. This anorectic or hypophagic effect was substantiated by a second trial in mice, which also showed that Δ9-THCV reduced body weight (34). As a caveat, a Δ9-THCV rich cannabis extract did not produce the same effect. The authors speculate that this was because of residual Δ9-THC, and were able to restore the effect by supplementing the extract with CBD. This demonstrates that patients seeking these therapeutic effects may need to try a variety of cannabis preparations. Although Riedel and colleagues (34) demonstrated these effects in lean mice, research has also indicated that Δ9-THCV can improve glucose tolerance in two murine obesity models and increase insulin sensitivity in the diet-induced model (35).

Δ9-THCV has also dramatically reduced pain sensitivity in two murine in vivo models (36). Notably, the animals’ average nociceptive response was decreased from 2 min to almost 0 in one model, and the withdrawal latency (time from stimulus to pain response) was increased about two-fold in both models. Δ9-THCV also reduced edema (swelling) by about 25% in both models. Δ9-THCV has also been tested for antiepileptic activity; in that study it reduced epileptiform activity in vitro by about half, and increased the number of animals with a complete absence of induced seizures (37). Taken together, these results indicate potential utility for Δ9-THCV in many human conditions.

**Cannabinoid Acids**

Certificates of analysis usually contain data about cannabinoid acid content as well as the cannabinoid they are derived from. Cannabinoid acids are believed to have largely the same effect as their activated forms, that is, Δ9-THCA has the same effects as Δ9-THC, because cannabionic acids are decarboxylated upon heating and lose their carboxylic acid moiety, as the creation of CO₂ is extremely favorable thermodynamically. This means that cannabinoid acids will be converted to their activated form when cannabis is smoked, vaporized, or heated,
Terpenes

Although 140 terpenes have been identified in cannabis, this review will focus on the 17 most abundant (Table I) (39). Except for Δ<sup>3</sup>-carene, all of these are designated as “Generally Recognized As Safe” (GRAS) by the FDA, or are approved as food additives by the Flavor and Extract Manufacturers’ Association (9). Therefore, medical cannabis patients can be relatively confident that selecting for desired terpene effects will not affect the safety of their medications. Although terpenes are not a major constituent of cannabis (less than 4% by weight), they are highly bioavailable through inhalation (reviewed in [9]). Most cannabis terpenes have been widely studied in animal models and in vitro to earn their safety designation.

Three terpenes with an overwhelming variety of documented effects are β-myrcene, D-limonene, and β-caryophyllene. There is evidence that β-myrcene can exert sedative, anti-inflammatory, analgesic, muscle-relaxant, and neuroprotective effects. It may also have potential applications in arthritis and peptic ulcers (9). Similarly, D-limonene has been reported to have sedative, antidepressant, antimicrobial, anticancer, analgesic, and anti-inflammatory effects, with therapeutic prospects for patients suffering from gastro-esophageal reflux, colitis, or obesity (9). β-caryophyllene, the most common terpenoid in cannabis extracts, has demonstrated evidence for anti-inflammatory, analgesic, anti-anxiety, antidepressant, cardioprotective, hepatoprotective, gastroprotective, neuroprotective, nephroprotective, antioxidant, antimicrobial, and immunomodulatory effects (9). As these three examples show, given the sheer number of animal and cell culture studies on terpenes, along with the challenges in using them to shape recommendations for medical cannabis patients, this review will address only those terpene studies involving humans, as well as some of the more general claims about cannabis terpenes (Table I).

Proposed Antioxidant Effects

Most cannabis terpene molecules contain one or more double bonds between carbon atoms (9). These double bonds can often act as electron-accepting centers, giving them antioxidant properties in vitro. Antioxidants are beneficial to organisms partly because they can scavenge these highly-reactive, damaging free radicals by directly accepting unpaired electrons (40). Thus, antioxidants have been associated with a reduction in the cellular damage that can lead to cancer (41,42). We can therefore expect that some cannabis terpenes might act as antioxidants and chemoprotective agents against cellular damage and cancer, and many of the terpenes have already been tested for these purposes (43–57); reviewed in (9).

Notably, antioxidant studies do not consistently support or refute the idea of antioxidant administration having clinical value (58–60). A more recent study suggested that administration of exogenous antioxidants could actually increase the likelihood of metastatic events in cancerous mice (61). Another unknown is how antioxidant properties of terpenes may affect medical cannabis patients differently depending on the route of administration, since part of the harm to cannabis smokers’ lungs is thought to be caused by oxidative stress (62). Because of the complications inherent in interpreting antioxidant properties, the following discussion will focus on observed physiological effects, rather than potential antioxidant effects of the most prevalent cannabis terpenes.

Proposed Antimicrobial Effects

It is widely understood that plants produce terpenes for protection against pests (63). Therefore, it is unsurprising that many terpenes have well-documented activity against bacterial (45,48,64–74), fungal (48,65,75–78), and protozoan pests (74,79). However, since these were all in vitro studies, it remains unclear how these antimicrobial effects might help prevent infections in medical cannabis smokers.

Proposed Neurobiological Effects

Proposed neurobiological effects for many cannabis terpenes include analgesic, sedative, anticonvulsant, anti-anxiety, and antidepressant properties (9). However, with few exceptions, these studies have mostly been undertaken in rodent models, making it impossible to extrapolate how different dosages or routes of administration might affect human...
patients. Additionally, since cannabis terpenes are typically consumed in combination with Δ9-THC, CBD, and other cannabinoids, it is difficult to predict how specific neurobiological effects of terpenes might interact with neurobiological effects from cannabinoids.

Human Studies

α-Pinene

α-Pinene has been studied in humans as an irritant and bronchodilator (80). Although it had mildly irritating effects, measures of bronchodilation were not significantly changed. In this study, irritation resulted from an atmospheric concentration of 450 mg/m³, a level unlikely to be found in cannabis smoke.

Δ3-Carene

Sweden has established the occupational exposure limit for Δ3-carene at 150 mg/m³ (81), given its documented effects as an eye and nose irritant (81,82). Similarly, this concentration of Δ3-carene is higher than in cannabis smoke.

D-Limone

As the main component in citrus fragrance, limonene has been studied in humans for treatment of depression (83). In this study, citrus fragrance reduced Hamilton Depression Scores (HADS) to a similar extent as tricyclic antidepressants, reduced urinary cortisol and dopamine levels, and reduced perceived need for antidepressants. Limonene has also been studied in Phase II clinical trials on advanced human breast cancer patients, but the treatment was ineffective (84). More promisingly, daily supplementation with a 95% limonene preparation reduced a peripheral inflammatory marker in elderly humans (85).

Linalool

A recent study retrospectively analyzed the anticonvulsant effects of cannabis in patients with epilepsy (86), and noted that strains higher in linalool may be more effective. However, retrospective observations such as these must be interpreted critically, as they lack rigor when compared with randomized, controlled trials.

Patient Summary

Every effort has been made to address the limitations of the animal and in vitro studies presented here. From the available data, no conclusions can be drawn on how any cannabinoid or terpene preparation, dosage, or route of administration will affect humans. The FDA has not approved even a single drug containing lesser-known cannabinoids for the treatment of any condition. Exaggerated claims about the cannabinoids and terpenes discussed above are rampant on the internet; leading sites feature blog articles that make claims about these phytochemicals apparently unsubstantiated by in vivo or in vitro evidence. This underscores the need for better patient and caregiver education on the phytochemical components of medical cannabis products. Furthermore, cannabis flower probably lacks sufficient concentrations of any of the minor

Figure 2: CBC-type compounds show variable antibacterial activity. (a) CBC molecule skeleton. "R" denotes where chemical analogs differ (90). (b)–(f). Diameter of bacterial inhibition zones 48 h after application of CBC compounds to Bacillus subtilis (b), Escherichia coli (c), Mycobacterium smegmatis (d), Pseudomonas aeruginosa (e), and Staphylococcus aureus (f) cultures (19).
cannabinoids to deliver their medicinal effects. For example, a recent online search found that the strains with the highest CBC content measured at only ~0.5% of dry weight. Similarly, CBC, CBG, and CBN are minor cannabinoids and most strains contain 1% or less, although CBN content can be increased with storage (87). Selective breeding or genetic modification might create CBC and CBG strains in the future, but patients seeking relief today may wish to try extracts or other processed products.

CBC, CBG, and CBN
Preparations of these cannabinoids show strong antibiotic activity. They may aid patients who are immunocompromised or otherwise at risk for infections. However, commercially available extraction products will have varying levels of activity based on the specific composition of different CBC and CBG analogues. CBG and CBN have the potential to ameliorate acne and psoriasis as they inhibit keratinocyte proliferation. The evidence is comparatively weak for their anti-inflammatory, antitumor, and analgesic effects. CBC and CBN may act to increase sedative effects of other drugs, but this result has not been replicated.

CBDV
Cannabidivarin has strong anticonvulsant properties in mice and rats, and a CBDV preparation has passed Phase I clinical trials. Epileptic patients who suffer side effects from CBD or incomplete reduction in seizures might consider trying CBDV.

Δ²-THCV
Single studies show that Δ²-THCV may aid in weight loss, ameliorate insulin resistance, reduce nausea, pain, and epileptic seizures, although CBD also has these therapeutic properties (88,89). Appetite reduction is the only physiological application of Δ²-THCV which has been reproducible.

Terpenes
Although terpenes are found in low concentrations in cannabinoids, they are highly bioavailable (9). Amongst the 17 most common cannabis terpenes, there is substantial overlap in predicted effects. For example, multiple terpenes have shown evidence of antioxidant, antimicrobial, analgesic, sedative, anticonvulsant, antianxiety, and antidepressant effects, however no human data is available for most of these effects. Add to this the low concentrations and variable terpene profiles in different cannabis strains, and it becomes extremely complicated to recommend specific terpene profiles for any specific purpose. Single human studies do support anticonvulsant and anti-inflammatory effects from limonene (83) and another suggests an anticonvulsant effect (86) from linalool. Therefore, patients seeking anticonvulsant and anti-inflammatory effects might experiment with high-limonene strains, and patients seeking anticonvulsant effects might experiment with high linalool strains under the supervision of their physicians.

Conclusion
Few recommendations can be drawn from existing research on terpenes and minor cannabinoids. As cannabis moves towards legality in the U.S., this should facilitate more rigorous human studies to help prescribers, budtenders, and medical cannabis patients obtain suitable medicines for their individual needs.

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References
Factors Influencing Yields in Extraction, Part II: Examining the Impact of Material Preparation

This three-part series began with Part I, an in-depth discussion of how the feedstock component makeup influences the yield obtained from an extraction. It examined starting material influences, including total mass of material loaded in the extraction chamber and the fraction of interest in the component makeup of the material. An overview of basic extraction mathematics with special focus on percent extraction efficiency calculations was also presented. Here, in Part II, we explore the impact of how material is prepared prior to extraction. To follow in the next issue, Part III will conclude the series with a discussion of the influence of total extraction time and processing parameters.

Krista Kulczycki and Aaron Godin

In addition to the influence of how much material is committed to a given extraction, and what fraction of that material represents component(s) of interest, decisions regarding material preparation prior to commencing the extraction also play a role in the yield obtained.

Particle Size
Specifically, we consider the role of feedstock particle size and its effects on packing efficiency and solvent-matrix interactions.

Packing Efficiency
Intuitively, reducing the feedstock particle size prior to its addition to the extraction chamber maximizes the mass of feedstock that can fit snugly into the space. In other words, in a fixed-volume chamber, and given the same feedstock material, more feedstock will fit into the volume of the chamber when ground to a finer size. One can equivalently consider trying to place rocks in a jar (Figure 1). Only a few large rocks can be maneuvered into a space of a given size. If the rocks are first pulverized, however, many more rocks’ worth of material can be fit into an identical space. Finer-sized material has better packing efficiency.

Time to Reach the Center of a Sphere
Further, reducing the size of the feedstock particle reduces the distance that must be traversed by the solvent to penetrate the centre of the particle (feedstock particle radius). Figure 2 considers a baseball diamond and two runners of identical speed. The first runner will start from the edge of the infield, the second from the edge of the outfield; both time a run straight to home plate. For reference, home plate is approximately twice as far from the outfield as it is from the infield. Which athlete is expected to complete their run in the shortest amount of time? For identical speeds of travel, a shorter travel time is required to traverse a shorter distance. In terms of extraction parameters, the solvent will reach the centre of a finer-ground feedstock particle more quickly than a coarser-ground one.

Surface-Area-to-Volume Ratio
Another factor for discussion with regards to particle size is the surface-area-to-volume ratio (SAV). Increasing the SAV increases interaction between the solvent and the feedstock matrix which, in turn, increases the rate at which the components of interest are brought into solution. This is easily the most complex of the effects to explain, as well as to internalize.

Figure 3 tabulates the radius, surface area, volume and SAV for each of three spheres, represented by three circles. As shown, a larger sphere has a larger radius, volume, and surface area. However, because these do not increase proportionally, the SAV actually decreases for larger spheres.

If one imagines hammering as many finishing nails as possible into a set of wooden spheres with dimensions as given in Figure 3, the smallest sphere would accommodate four times as many nails, compared to its volume, as the largest sphere. The largest sphere would still have more nails, but its volume is much less accessible.

Each point on the surface of the sphere acts as an avenue of interaction with the sphere’s volume. A smaller particle size provides more avenues of interaction per particle volume than does a larger particle size. This allows interaction of the solvent with the matrix of the feedstock material to occur more efficiently.

Coextraction of Other Components
To round out the present discussion of the merits of reducing particle size, it is important to note one particular point...
regarding extraction without particle size reduction. The process of reducing particle size also disrupts plant cell structure. Rupturing cells provides increased access to the interior contents of the cell, some of which may not be desired in the end product. Thus, while the advantages of smaller particle size offer increased efficiency in extraction, it can also mean more coextraction of other components. Extraction without particle size reduction can provide access to desirable components while employing the plant’s own structure to help minimize coextraction.

Trade-offs in total extraction time, extraction efficiency and post-processing requirements thus have many layers of complexity as an operator prepares for an extraction. The choice of extraction parameters, discussed in the next part of this series, provides the operator with additional tools as they work to identify the most desirable pre-processing, extraction, and post-processing regime.

Conclusions

In summary, a smaller particle size provides two main efficiency increases: an advantage in packing efficiency, and an increased efficiency of interaction of the solvent and feedstock matrix through reduced particle radius and improved surface-area-to-volume ratio. The process of reducing particle size may also increase coextraction of potentially undesirable components. These concepts, with those discussed in the series’ first installment, conclude the discussion of starting-material influence on yields. Part III, the next and final in the series, will examine the effects of processing parameters on an extraction’s output.

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High-Resolution Tandem MS Applications in Cannabis Product Development

As cannabis legalization gains momentum, an emerging industry has been created that has a need for accurate, reliable and innovative analytical testing. In my role as Director of Analytical Chemistry at Next Frontier Biosciences, I chose high-resolution liquid chromatography/mass spectrometry (LC/MS) to meet these needs. In this article I will discuss applications in routine testing (potency and contaminants) as well as those more specialized (terpenes, lipids, etc.). Tandem quadrupole/ time-of-flight (QTOF) mass spectrometry is shown to be a valuable tool in routine and custom applications that will contribute to the next generation of cannabis products. LC/MS capability in the cannabis laboratory goes well beyond contaminant testing currently used in most cannabis third-party testing labs.

Kris Chupka

Cannabis legalization has created a new industry for consumer products with recreational and medical applications. The diversity and complexity of products in development rely on thorough and accurate characterization to meet desired endpoints. Often developers use third-party laboratories to perform this characterization. Mass spectrometry is a key technology for contaminant testing but is often overlooked for other applications. In this article I will recount my time as Director of Analytical for a cannabis biotech company and the high resolution-mass spectrometry (HRMS) applications we developed and used for our research and product development.

Quantitative Cannabinoid Potency by LC/QTOF

In the cannabis industry, the most immediate need for characterization is potency of cannabinoids in flower and concentrates. HRMS was selected as our primary analytical technology so I initially worked to develop a separation and quantification for cannabinoids.

It’s worth mentioning that HRMS does not provide inherent selectivity for cannabinoids with the same molecular formula. Our scientists evaluated this first-hand and found the differences in MS/MS were too subtle for routine use in quantitation. The necessity for a good LC separation was imperative. Our initial efforts focused on identifying a column stationary phase that had unique selectivity for cannabinoids. The screening process was evaluated by resolution of cannabinoids (CBD), Delta 9 tetrahydrocannabinol (Δ9-THC) and Delta 8 tetrahydrocannabinol (Δ8-THC) under isocratic conditions. Our laboratory screened more than 10 phases from several manufacturers. Once the phase providing the greatest resolution was identified, we worked on other parameters to obtain a separation of 12 cannabinoids. An example is shown in Figure 1 of the signals for m/z 315.232 and 359.222.

Not long after our separation was established we found that, for accurate quantitation in the various matrices, there was a need for a relevant internal standard (ISTD). The evolution of ISTD in our laboratory started with deuterated cannabinoids, a paraben, and finally an isoflavone. Unfortunately, we found the purity of the deuterated standards available at the time less than adequate and the paraben was prolific in the environment.

The quality of cannabinoid standards has improved greatly since that time, so these may be worth re-evaluating. However for our application, the isoflavone worked well. We performed basic validation (accuracy, linearity selectivity) in flower and concentrate and comparison studies with liquid chromatography/ultraviolet detection (LC/UV). The results of our equivalency study is shown in Figure 2.

Our accuracy for cannabinoids was routinely ±10% or better (as monitored by check standards and a control sample). While this may not rival the best LC/UV techniques, we had some inherent advantages with HRMS. The advantages of using HRMS observed were selectivity for cannabinoids versus matrix and the ability to run information-dependent MS/MS scans information dependent acquisition (IDA) in the background for identification of unknown components of interest or verification of compounds identified. The IDA data could be re-evaluated weeks or months after a sample was tested to look for non-target components of...
interest. Our laboratory analysed hundreds of flower and oil samples with a high degree of confidence in the results.

**Semi-Quantitative Terpenes by LC/QTOF**

After successful implementation of a quantitative cannabinoid HRMS analytical method, the next obvious need was characterization of terpenes. LC/MS is not typically thought of for analysis of volatile compounds but has some advantages over thermally intensive methods such as gas chromatography (GC). The initial work in my laboratory was to simply obtain terpenes response in the MS. This was accomplished through atmospheric pressure chemical ionization (APCI). Again the common molecular formula pointed to the need for good chromatographic resolution.

Unfortunately, our laboratory equipped our LC/MS with a high-performance liquid chromatography (HPLC) instead of an ultra-performance liquid chromatography (UPLC). There were significant limitations in the resolution of terpenes with a standard HPLC method. As demonstrated for other complex separations, UPLC probably would be a better choice. However, for our research the partial resolution and semi-quantitative information was sufficient to characterize the samples of interest. We could easily distinguish classes of terpenes (monoterpenes versus sesquiterpenes) and differentiate strains of cannabis. Figure 3 shows an example of terpene standard analysed by HRMS with identification of common terpenes.

**Cannabis Lipidomics**

Due to potential impact on finished product quality and customer appeal, lipid content of
cannabis materials quickly drew the attention of our organization. Our raw material suppliers frequently described a myriad of processes to remove lipids. Lipids were of known concern to our formulators and little was known about what species were present or how effective the removal processes were. For this project, our lab utilized lipidomics (Sciex Lipidview) software and HRMS to create profiles of lipids in cannabis oils. The application was almost exactly that used in biological lipid characterization where an extract is infused into the MS and known parent lipid m/z and fragments are measured. Figure 4 shows the negative ion fatty acid lipid profile of hemp oil. Figure 5 shows the positive ion triacyl glyceride profile of hemp oil. Hemp oil was chosen for development as it would be easy to implement as a long-term control sample.

Our laboratory was successful in providing semi-quantitative values for lipid species and to characterize effectiveness of lipid removal processes. It was speculated this data could be useful in developing IP lipid removal processes and determining if the lipids themselves may have commercial value. Unfortunately our laboratory wasn’t able to perform widespread characterization of cannabis materials using this technology, but we were able to demonstrate feasibility.
Conclusion

The complexity of the cannabis plant coupled with a diverse emerging commercial market provides significant opportunity for analytical scientists to utilize HRMS beyond contaminate testing. The applications I have described here only serve to highlight the utility of HRMS in this industry.

My hope is contract laboratories and cannabis product development companies can rethink initial capital expenditure in LC/MS to obtain improved return on these investments.

Figure 5: Positive ion triacyl glyceride profile of hemp oil.

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How ERP Solutions Support Accountability in Cannabis Businesses

As legalization expands and regulations change, an integrated enterprise resource planning (ERP) solution developed for growers or manufacturers of cannabis products goes beyond the basics of accounting and manufacturing to provide control and visibility over data, inventory, quality, and traceability. With evolving government regulations and scrutiny from diversified consumers, this industry is driven to maintain stringent accountability for compliance to establish and remain operational. ERP software supports a detailed level of seed-to-sale traceability by providing lot and plant tracking for regulatory compliance, real-time inventory tracking, and in-depth reporting and analysis to meet the increasing pressures of this industry. Also faced with evolving quality control demands in the edibles marketplace, established manufacturers are finding an ERP’s tools essential, including research and development, established formula and recipe management, and recall preparedness plans. Ultimately, a business management solution versed in meeting the compliance and regulatory needs of processors in highly regulated industries is becoming a sought-after asset for those who are willing to commit. This article dives into three specific areas in how ERP solutions support accountability in cannabusiness: traceability and compliance; established inventory control measures; and government reporting requirements. The article also informs the reader about what’s next with edibles regulations.

Daniel Erickson

The expansion of marijuana legalization and the subsequent ever-changing environment brings forth challenges and complexities for cannabis cultivators, manufacturers, processors and dispensaries looking for effective business management solutions to meet today’s market demands. With evolving government regulations and scrutiny from savvy customers, it’s important for cannabis operations to have systems in place to handle more than just the basics of accounting and manufacturing. Proactive cannabis businesses are seeking technological solutions to provide the control and visibility required for traceability, compliance, inventory control, governmental reporting and food safety. An integrated enterprise resource planning (ERP) solution developed for this unique industry allows companies to maintain stringent accountability for compliance while streamlining processes and preserving the transparency necessary to navigate the complexities of the cannabis marketplace.

The evolving nature of marijuana laws and regulations are the hallmarks of the cannabis industry in its beginnings of legitimacy. While emerging as a recognized industry, cannabis industry lawmakers and regulators are acknowledging that their business needs resemble other process manufacturing verticals that benefit from ERP system solutions such as dietary supplement, pharmaceutical, and food and beverage companies. ERP software packages are an increasingly popular choice for established cannabis companies seeking to capitalize on the processes and industry-best practices already in place for other highly regulated industries to maintain a competitive advantage.

Similar to other regulated industries, cannabis operations require a focus on traceability, inventory and process control, accountability and finances that includes dynamic reporting and documentation. ERP software supports a detailed level of seed-to-sale traceability by providing real-time lot and plant tracking through greenhouse management, inventory transaction recording and in-depth reporting and analysis. Also faced with evolving quality control demands in the edibles marketplace, manufacturers are finding ERP tools essential, including current good manufacturing practices (CGMP), research and development, formula and recipe management and recall preparedness plans. Ultimately, a business management solution versed in meeting the compliance and regulatory needs of
processors in highly regulated industries is becoming a sought-after technological asset.

With or without federal regulations, cannabis enthusiasts have an expectation that the products they purchase for consumption are consistent and safe. It is necessary for marijuana processors and distributors to embrace this fact.

Historically, the cannabis industry has received an unfavorable reputation for producing inconsistent products, which results in loss of consumer trust, and is detrimental to the success of the industry—specifically the edibles niche. There is a strong possibility for these types of companies to create consumer perception problems that cause disruptions in the market and directly impact the success of similar businesses, creating a bad reputation for cannabis products overall. With the lack of legal regulations on the federal level in the United States, the industry itself is pushing for standardized guidelines that focus on the production of consistent, high-quality products. Integrating business operations and compliance reporting provides accountability features that are effectively handled with an ERP solution.

In an effort to promote quality standards and processes, many cannabis companies have implemented CGMPs, prerequisite programs (foundation of hazard analysis critical control points [HACCP]), global food safety initiative (GFSI) certified practices and food safety guidelines that mainstream food and beverage manufacturers already abide by. Applying best practices regarding formula and recipe management, plus quality and product labeling, enables operations to manage security, revision control and meet label claims, especially in regards to cannabinoid (CBD) and tetrahydrocannabinol (THC) levels. An ERP solution supports following of proper procedures and automating documentation to efficiently address food safety concerns.

**ERP Solutions Provide Accountability**

With an ERP system, every transaction is fully integrated with real-time visibility across all departments of a business—a reality not possible with disparate systems or paper spreadsheets. Automation, recording and streamlining of critical business functions such as cultivation, purchasing, production, quality, inventory, costing, sales, financials and reporting provide the accountability needed in order to improve operations and meet regulatory requirements. A fully integrated ERP solution is adaptable and scalable in order to handle business growth, including addition of product lines and locations, legislative changes and market trends—providing a comprehensive set of tools with the ability to incorporate additional features as needed.

In this emerging industry, software providers with in-depth experience regarding regulatory processes and audit procedures are enabling businesses to maintain oversight and adapt to trends that will inevitably develop in the future. A vendor with an experienced background in dietary supplement and food and beverage manufacturing or other similarly regulated industries employs a breadth of knowledge and a solid foundation for cannabis ERP software without the need to reinvent the wheel. Building on this experience, cannabis ERP providers tailor the platform to meet the needs of the constantly growing cannabis industry.

The absence of readily available financial institutional resources for this industry has placed emphasis on utilizing sound accounting solutions integrated in ERP systems to provide detailed and transparent bookkeeping for an often cash-only operation. With full accounting and general ledger control, companies have a handle on every transaction with proper documentation to avoid legal and regulatory penalties. Comprehensive reporting and analytics functionality provides interactive visualization in key areas of business. With the ability to automate control, budgeting and forecasting, the real-time information available in the financial reporting tools gives cannabis companies the information needed to handle the complex reality of the industry and improve their bottom line.

**How ERP Solutions Support Accountability in Cannabis Businesses**

**Traceability and Compliance**

Addressing regulatory and compliance issues is a primary concern in the industry, as more U.S. states are legalizing medical and recreational marijuana while Canada’s federally legal cannabis market continues to grow. With each jurisdiction employing its own guidelines and rules, the potential for confusion and law interpretation differences are a strong possibility. Preparation and planning to ensure compliance with new and evolving regulations is a necessity to avoid fines, product recalls, product seizures and other penalties.

The emphasis on traceability is attributed to the Cole Memo which was drafted by former U.S. Deputy Attorney General James Cole, providing guidance for municipalities that have legalized medicinal and recreational marijuana. The memo directs federal legal resources away from prosecution of operations that are in compliance with the local jurisdictions for which it has been legalized. It encourages companies to implement strict inventory control and transparent tracking documentation to avoid...
enforcement of federal law. In early 2018, U. S. Attorney General Jeff Sessions rescinded the memo, and little has changed in regards to companies following its guidelines in the areas of traceability and compliance.

ERP solutions with cultivation management and other supporting functionality provide a centralized framework for forward and backward lot traceability and unique plant ID tracking of marijuana and its derivatives. No matter the stage of the marijuana plant or byproduct, best practices are utilized to provide an audit trail for all greenhouse activities, encompassing tracking, measuring, documenting and reporting from seed-to-sale. Similar to other manufacturing industries, product recall capabilities within an industry-specific ERP solution are essential to keep consumers safe from inconsistent, unsafe or contaminated products. An ERP system streamlines the maintenance of key requirements, traceability, waste disposal, tax payments, customer and product tracking, packaging, labeling and transportation to address the multiple and competing compliance demands as well as jurisdictional changes.

Established Inventory Control Measures
Cultivation management functionality within an ERP tracks and manages each individual plant through the growth stages with a unique identifier that goes beyond merely assigning a plant ID—monitoring genealogy, mother and cloning, CBD and THC content, labor costs and quality control checks throughout the cultivation cycle. Tracing products back to the strain or variety is equally important, documenting the health, weight and optimal growing conditions of plants and groups of plants, accurately recording of the entire lifecycle. Performance of strains and their receipt by the marketplace are recorded, helping to determine the ideal market for selling and distributing a company’s cannabis products. User-based permissions within an ERP solution ensure the security and accountability of operations by allowing designated personnel to perform transactions and access data.

With plant growth cycles of 2-3 months, cultivation management is a necessary component of tracking growing activities, inventory and labor needs throughout greenhouse operations. Due to the extended growing period, there is a multitude of data and expenses that need to be tracked to manage costs and ensure quality control—assessing plant health, movement, growth stage and harvest information. With the highest greenhouse costs related to labor, continuous management and monitoring is essential. Manual processes lack the accuracy, transparency and security necessary to effectively manage cultivation stages.

Established inventory control measures such as stock levels, expiration dates and loss tracking are managed in the ERP solution, assisting processors in maintaining appropriate levels, reducing waste, utilizing rotation methods and avoiding over-production to help control overhead costs. To minimize the risk of unsafe products entering the market, the real-time and integrated information available in an ERP solution facilitates the process of locating items quickly in the event of contamination or recall.

Government Reporting Requirements
Reporting capabilities to governing bodies is a must-have, and a comprehensive ERP has the strict inventory control, traceability and chain-of-custody documentation to ensure compliance. Whether reporting is required to state municipalities or at the federal level (as seen in Canadian operations), integration to approved regulatory compliance systems is essential. With differences in jurisdictional requirements, it is important to implement a system that compiles real-time company data and effectively produces timely reports to adhere to the rules, regulations and statutes of the particular jurisdiction. Failure to do so can lead to criminal penalties and loss of license, both of which are detrimental to a company’s success. An ERP solution that integrates and communicates with approved traceability systems streamlines compliance procedures and ensures accountability at all levels within the organization.

Cannabis Edibles
Consumer expectations for product and food safety are as equally important to account for in the edibles market as they are in the mainstream food and beverage market. With the cannabis edibles and infused beverage market increasing in popularity for both medicinal and recreational use, it is necessary for manufacturers to address food safety despite the lack of federal legality and regulatory guidelines.

Forward-thinking cannabis operations have realized that following current food safety guidelines, incorporating best practices and implementing an industry-specific ERP solution to automate processes provides the accountability they need to ensure safe, consistent and high-quality consumable products.

Foodborne illness due to unsafe and unsanitary manufacturing processes is an increasing possibility, as U. S. Food and Drug Administration (FDA) guidelines for food and beverage products sold in the United States are not mandatorily enforced in the cannabis-infused marketplace. Without established federal standards in
this industry, product and food safety risks to customers are not monitored, documented or reported. Cannabis-specific concerns regarding aflatoxins, plant pesticide residue, pest contamination and improper employee procedures and training heighten the opportunities for risk. Inconsistent levels of THC and CBD potency in consumables can also be added to the list of threats that can lead to recalls, monetary expenses and damaged reputations.

By focusing on quality and safety, proactive edibles producers are investing in ERP solutions with vendors experienced in food safety in order to take advantage of the benefits traditional food and beverage manufacturers have experienced for decades. Standardizing and documenting procedures for your company and implementing an ERP solution includes employing CGMPs to ensure safe and sanitary manufacturing, storage and packaging of products. In addition, establishing a food safety team to satisfy HACCP requirements is essential. Instilling these safeguards protects consumers from biological, chemical and physical dangers of cannabis edibles and infused beverages. A documented food safety plan (FSP) ensures that food safety policies and procedures are followed. In addition, ERP functionalities such as inventory control; recipe and formulation management; lot, batch and plant tracking; and accurate product labeling, further support control within the supply chain, food safety initiatives and quality control measures.

The evolving cannabis industry faces accountability challenges on a daily basis and an industry-specific ERP system can provide the business management solution to navigate the needs of this unique industry—providing traceability and compliance, inventory control, integrated governmental reporting and the handling of food safety challenges. By employing an ERP vendor that keeps up with current regulations, as well as future changes as legalization continues, companies can meet demands, maintain compliance, differentiate from the competition and allow for operations to grow profitably.

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