The Use of Near Infrared Spectroscopy for Potency Measurement

*Introduction to the Luminary™ Profiler*

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I. Background

a. State of Cannabis Industry

The evolution of the cannabis industry in the U.S. has been far from ordinary. Marijuana currently remains a Schedule I drug under the federal Controlled Substances Act of 1970, and thus illegal for any reason under federal laws, with the exception of FDA-approved research programs. Notwithstanding, states have taken steps to legalize the substance at a slow and steady rate since 1998, when voters passed approval of medicinal marijuana, and in the past five years, it has accelerated to 35 states having some form of legalization. As of November 2014, four states and the District of Columbia have legalized “adult use” or recreational marijuana, all through voter initiatives as opposed to legislative mandate.

Because of this slow and awkward growth of the quasi-legal market for marijuana, complementary markets have been understandably reluctant to organize. For this reason, development of the testing technologies focused on the applications of potency and safety measurements of cannabis have been relatively stagnant.

b. Why is Potency Measurement Needed?

Potency, in the traditional pharmaceutical sense, means the amount of active ingredient in a product. In cannabis, potency is driven by tetrahydrocannabinolic acid, THC, which is its primary and most prevalent psychoactive compound. In flowering products, THC content is generally around 10 to 20% by weight. In hash and extracted oils, this content can be much higher (up to 80% or more by weight). The second element of cannabis potency is cannabidiol, CBD, which does not provide the psychoactive stimulation of THC but, rather, physiological relaxation associated with therapeutic usage of cannabis (anxiety relief, muscle relaxation, appetite stimulation, among others). CBD is typically present in much smaller concentrations, less than 10% by weight and usually closer to 1 or 2%. The third component of potency is cannabinol, CBN, which is a very weak psychoactive compound and, importantly, a natural breakdown product of THC that occurs over time. In fresh product, CBN is typically at or near 0, but this percentage increases as the product slowly degrades over time. As such, CBN can be a useful indicator of the product freshness. While multiple other cannabinoids are present in cannabis, these three are the most widely studied and ascribed physiologic importance. Future scientific studies may reveal significant effects from the remaining cannabinoids, in which case their inclusion in the overall measurement of “potency” will be considered.

There are thousands of unique strains of cannabis being grown and consumed, yet the ability to present an accurate potency measurement for each has been severely limited due to the costly, and relatively inaccessible methods that have been available for strain testing. Consequently, marijuana
users have not been assured of receiving an accurately labeled product, or of product consistency over multiple purchases. While frustrating for recreational users, it can mean devastation for medical marijuana patients whose abatement of painful symptoms depends on the critical need for both accuracy and consistency.

c. State of the Art in Cannabis Measurement

The state of the art in cannabis chemical characterization entails established laboratory analytical techniques, including gas and liquid chromatography (GC and LC) and mass spectrometry methods. While these methods provide accurate molecular characterization and quantification, there are many drawbacks that impede its comprehensive adoption, and prevent it from enhancing the productivity of the rather unique nature of the cannabis industry. For example, GC and LC methods typically require sample preparation and destruction, the use of consumable preparatory materials, a skilled operator, and/or a waiting period of one to more typically several or more minutes to obtain a result. This timeframe is incorporated into the overall several days that it takes a grower, dispensary or regulator to ship the samples and await the results from the remote testing facilities that provide potency data. Further, hazardous waste is often a by-product of GC and LC testing methods. To date, it has been necessary for cannabis samples to be sent to dedicated laboratories, a relatively expensive and time consuming process, thus creating a bottleneck to optimal productivity of cannabis industry operations.

Because of these impediments, regulators have only required a small sample or a larger lot to be actually tested. However, due to the natural variations inherent in such an organic plant substance as cannabis, the true potency of each product of the lot that gets consumed but was not actually tested cannot be known for certain by the user.

It is important for customers to have accurate potency measurements of the medical and recreational cannabis products they are purchasing. Additionally, the still nascent regulatory climate is maturing rapidly and will predictably reach the levels of stringency required for alcohol and pharmaceuticals. Testing procedures that are more flexible, practical and affordable will facilitate these needs and developments.
II. Why is Optical Spectroscopy the Preferred Solution for Cannabis Potency Testing?

Optical techniques such as reflectance and absorption possess several hallmark advantages over conventional analytical techniques for chemical characterization and quantification. Optical methods are nonintrusive to the sample, which allows measurement without altering the chemical content or causing any physical change to the sample. Other benefits of optical spectroscopy include: allowing rapid sample measurements (typically in less than one second), molecular specificity, such that individual compounds of interest can be identified distinctively, typically without any further sample preparation, automated characterization or quantification, allowing their use by unskilled personnel or with minimum training, portability for use on-site and with as much frequency as desired, and lastly, use of devices that require little or no maintenance over their operating lifetime.

a. What is Near Infrared (NIR) light?

Visible light, which ranges from about 400 to 800 nm, contains almost no chemical information and is actually a measure of a material’s color. Near-infrared (NIR) light is the region of the electromagnetic spectrum just beyond the red light that our eyes can see, around 800 nanometers (nm), and extends to approximately 2,500 nm. This wavelength range has become increasingly popular for rapid chemical assessment in a number of industries, because it contains a wealth of information and can be measured very quickly. Light in this region of the spectrum interacts with the chemical bonds in molecules. By measuring the light intensity returned from the samples at each point in the spectrum, characteristic fingerprints can be accurately and rapidly measured. NIR can be used to monitor both chemical and physical properties, and has been frequently employed for qualitative analyses in the food, chemical, oil, gas, petrochemical, feed, agriculture and pharmaceutical industries.

b. Introduction to the LuminaryTM Profiler

The LuminaryTM Profiler developed by Sage Analytics (sageanalytics.com) uses the most information-rich portion of the NIR range (~1500 to 2000 nm), where chemical features are significantly more pronounced and immune to unimportant factors like the cannabis color. This enables a more precise measurement of the cannabinoids and product moisture. Diving deeper into the science, the NIR spectral region from approximately 700 to 2500 nm corresponds to overtones and combination bands of molecular vibrational absorption. The functional groups probed by NIR are predominantly those containing hydrogen bonds. Compared to mid-infrared spectroscopy, where fundamental absorptions are evaluated, NIR can be used to measure thick samples with high water content, and its allowance for diffuse scattering permits its use in both transmission and reflectance geometries, vastly simplifying sample preparation and measurement.
While NIR light is becoming more prevalent in various industries, the key enabler to its use is the ability to translate the NIR information into a meaningful value. In this case, the meaningful values are the cannabinoid quantities present in tested samples. Just as they have done for determining the exact amount of active drugs in pharmaceutical tablets as they are being produced, Sage Analytics’ scientists first characterized the NIR signature of the purified cannabinoids. This was done through a unique agreement with leading academic researchers, as such substances require precise scientific procedure, knowledge and institutional support to develop. From these cannabinoid signatures, sophisticated mathematical models were developed to accurately quantify the amount of each specific cannabinoid in the total NIR signal recorded from a tested product, regardless of product strain or type (or color). This is the same approach approved by the FDA for NIR use in pharmaceutical testing, and is a robust method to test any form of product.

Cannabis is a natural product, however, and like all natural products the variation is enormous: regions of the same bud will have different potency, as will different buds from the same plant, buds harvested at different times, buds stored for different amounts of time, etc. Since the Luminary™ Profiler only measures the NIR light signature of the cannabinoids however, none of this variation affects its measurement. The Luminary™ Cloud is used to access the set of mathematical formulas that translate the new sample’s light signature into accurate cannabinoid potency metrics. These algorithms are stored in the cloud so they can be continually improved, and to add further test parameters (e.g. additional cannabinoids and chemicals) as the Sage scientists hone their testing accuracy in their controlled laboratories with rigorous procedures, not using the samples sent in by users.

It is important to note that NIR spectroscopy, as employed in the Luminary™ Profiler, is considered a secondary analytical technique, whereas GC and LC are primary methods for evaluating cannabinoid potency. This simply means that the Luminary™ Profiler is reliant on a cannabinoid potency data set that has been previously characterized using a highly accurate, primary method for the development of models capable of calculating potency.

c. Why is NIR, and the Luminary™ Profiler, preferred for potency measurement?

Each participant of the cannabis market eco-system (growers, dispensaries, regulators, marijuana-infused product manufacturers) has an important need to ensure accuracy and consistency of their cannabis potency measurements, this necessity has been hindered by the inherent obstacles of, and omissions to current testing methods. Specifically, until now, nearly all testing must be conducted off-site of a business or regulators’ operations. This burden causes delay, and because of this delay, one’s operations are prone to error or inconsistency. Safety is an issue, as the need to transport marijuana samples having such a high monetary and black market value can present a risk to the transport agent. The sampling of cannabis plants in the field can lead to contamination of the samples to be evaluated, which can result in errors in the analysis. This contamination may stem from, for example, extraneous
debris adhering to the harvested plant material. Most notably, if on-site testing was available on a cost-effective basis, this would enable quality control and production efficiencies to increase considerably. Additionally, costs, delay and transport risks could be significantly reduced.

The Luminary™ Profiler enables potency measurements in a diverse assortment of cannabis-based products including flowers, kief, hash, oil, waxes, concentrates, etc. The majority of these products require no sample preparation, significantly decreasing experimental time and cost. The samples can be evaluated more rapidly, allowing a greater quantity of samples to be measured. This feature of the Luminary™ Profiler translates to the proficiency to analyze a greater proportion of available samples, as opposed to current methodologies, where a sample subset is deemed representative of the whole. This means that vast quantities of potential products are not independently analyzed due to the cost of such an endeavor. By analyzing more samples in less time, the Luminary Profiler provides the potential to exhaustively assess cannabinoid potency in all samples, leading to more detailed knowledge of the chemical composition of a cannabis strain, and therefore, more accurate labeling. Further, use of the Luminary Profiler enables existing GC/LC equipment at laboratories to be freed up to increase throughput of contaminant testing.

d. FDA Guidance Supporting Optical Technology for Process Analytics and Testing “PAT”

In 2004 guidance was provided by the Food and Drug Administration (FDA) to the pharmaceutical industry, titled “Guidance of Industry, PAT (Process Analytical Technology) – A Framework for Innovative Pharmaceutical Development, Manufacturing, and Quality Assurance”. This initiative has helped process improvements evolve in an otherwise risk-averse and reluctant industry and governing regulatory body. As changes to the testing and regulatory processes in the pharmaceutical industry were stymied from fear of process deviations, the FDA recognized it was missing opportunities for technological improvements, and thus implemented instructions to facilitate innovation and technological advances to enable production process improvements. The FDA guidance was warmly received, and was successful in increasing innovation of PAT including the use of NIR for analytical processes. NIR is the PAT instrument of choice encouraged by the FDA. Since the PAT initiative was unveiled over ten years ago, the advances that have been enabled are credited with causing a substantial decrease in the time required to bring drugs to market, significantly improving quality control in manufacturing, and cost savings throughout the pharmaceutical industry.

As the cannabis industry evolves through the legalization process, the coordination of its regulation follows. Indeed, the need for standardization throughout the marijuana industry is dire, as states are dealing with regulatory issues independently and in serial fashion, without a federal agency to oversee the efforts. Needs addressed in the FDA PAT Guidance include advances in process analyzers to make real-time control and quality assurance during manufacturing feasible (as opposed to off-line and remote testing); avoiding the need for sample preparation; and for integrated systems. Goals include
improving the scientific basis for regulatory specifications; promoting continuous improvement to ensure precision and consistency of production; and improving manufacturing while maintaining or improving current levels of production growth. This guidance is highly relevant to the similar needs and goals of the cannabis industry, and thus should be considered as a precursor to the regulatory guidance and rules that will inevitably continue to refine and evolve to ensure the safety of the public.

e. Industry benefits of NIR

There are many benefits that NIR spectroscopy, as used in the Luminary™ Profiler, can provide over traditional GC and LC techniques, and that are specific to the unique requirements of the evolving cannabis industry. NIR benefits include:

- Simplifies obtaining measurements of potency and moisture through highly accurate THC, CBD and CBN authentication at each point in the cannabis growing, production, regulation, distribution and sales ecosystem, without requiring a trained technician to process the testing and results.

- Provides a portable, on-site device for use by nearly every business in the cannabis supply chain.

- Provides in-process and instantaneous, highly accurate potency and freshness profiles for flowers, and potencies of concentrates and infused products, that can be viewed on a display screen or printed on a label or receipt.

- Are easily operated by any employee or customer; does not require a skilled operator.

- Rapid acquisition of data as opposed to standard LC and GC methods.

- Does not chemically alter the sample to be analyzed.

- Tests the samples without causing their destruction.

- Does not produce any hazardous waste byproduct.

- Retains the measurement results for a company’s sole access for use in quality control and archive purposes.

- Reduces the cost and complexities associated with remote lab testing by ensuring operations, and samples therefrom, are in optimal conformance with regulatory testing.

- On-line attributes reduce the risk of contamination through harvesting and transporting samples from the field to the lab.

- Increases safety of operations by requiring less transport of samples.

- Better flower or plant averaging by acquiring data from multiple plants and portions thereof, resulting in more realistic potency measurements.

- Greater accessibility to consumers by providing the ability to test and measure each product
purchase, thus increasing consumer safety through greater accuracy for both Adult Use customers and Medical Marijuana patients.

As applied to the cannabis industry ecosystem, the benefits attained from the Luminary™ Profiler products extend across most participants:

- Regulators and Government Officials can test instantly, on-site, and on their own, for active ingredient levels of THC, CBD, and CBN without relying on delays associated with remote lab testing – inexpensive, accurate testing is now available at their fingertips.

- Growers may regularly monitor potency of their crops to determine the optimal time to harvest, resulting in enhanced quality and consistency of the product. Potency levels of the flower may be verified while it remains on the plant.

- Laboratories are able to increase testing yields, significantly reducing the testing turn around time, and lowering the skill requirements for operators to measure THC, CBD, and/or CBN levels.
  - Existing labs will increase their businesses through these higher-throughput tools over existing, more complex GC and LC equipment (which can be used for other types of testing such as for contaminants).
  - Lower cost of entry and skill requirements enable more labs to be formed.

- Manufacturers of edibles and infused products can test potencies of concentrates and butters before processing them to assure desired and consistent cannabinoid levels in every batch.

- Medical marijuana dispensaries can offer a faster and less costly, yet accurate and reliable method to provide optimal THC and CBD ratios for their patients, at their facilities, and at the point of sale.

- Dispensaries have real-time point-of-sale measurements that enable their retail customers to have the assurance that the potency and freshness levels in flowers and extracts are accurate -- providing them confidence in the purchase of the product.
  - Product labels or receipts can be printed instantly upon testing while observed by the customer / recipient.

- Customers obtain more consistent and precise potency products.
III. Sampling

A major limitation of traditional analytical tools for evaluating cannabis potency is that the plant samples must be subjected to an extraction to remove the cannabinoids from the complex chemical matrix that composes the whole plant. These extractions often employ solvents, such as chloroform and methanol, which are health hazards, and require distinct safety precautions to be implemented. Additionally, if GC is used to measure potencies, the molecules must be volatile (meaning readily converted from a liquid to gaseous state). This means that the acidic forms of THC and CBD cannot be measured with GC, unless they are derivatized with a molecule that makes them more volatile. This added sample preparation requirement decreases the number of samples a researcher can measure in time. As previously mentioned, GC destroys the sample, and retrieval of the compounds of interest in LC would require costly and laborious purification.

The facility of the sample preparation using the Luminary™ Profiler enables researchers to spend more time evaluating samples, rather than time-consuming, potentially toxic preparatory steps. The Luminary Profiler can measure samples in any form (i.e., solids, oils, waxes). Whole buds can be used, however Sage Analytics’ scientists recommend that the samples be homogenized through grinding, to provide a more accurate and realistic measurement of cannabinoid levels throughout the bud. For example, trichomes contain high THC levels, and if the user honed in on this area of the plant, falsely high THC contents may be reported. Likewise, if the sample contains stem material, and the NIR light probes this region of the sample, anticipated cannabinoid levels will be skewed.
IV. Applications

a. Using the Luminary™ Profiler

The ease of use is one of the foremost features of the Luminary™ Profiler. Each system includes a detailed step-by-step software wizard to ensure that users feel comfortable in operating the instrument. A list of the main steps in acquiring cannabinoid potencies using this system is provided below:

- **Calibration**
  - When initially turning the system on, the Luminary Profiler requires a 20-minute warm up period to ensure that the lamp reaches a stable operating temperature. The system need only be turned off when it will not be used in order to save lifetime of the bulb.
  - The measurement window must be thoroughly cleaned with isopropyl (rubbing) alcohol wipes. The alcohol should be allowed sufficient time to completely evaporate, as it in moist form can potentially interfere with the potency analysis.
  - After cleaning the window, the system must be calibrated using two references, entitled “black” and “white” calibrations. These calibrations steps are mandatory when the Luminary Profiler is first powered on, and every 24 hours thereafter (once daily). They do not need to be performed before measuring every sample.
  - The black calibration cap is placed over the cleaned window. This provides a measurement of any signal produced by the instrument in the absence of NIR light and any samples.
  - The white calibration uses a National Institute of Standards and Technology Spectralon® reference puck. This is a completely reflective surface, and as the black calibration provided a lower limit of the spectral signal, the white reference provides the upper limit of a maximum amount of light coming into the measurement window. The white reference puck is encapsulated in glass, and therefore, should not be touched or scratched. The puck should be thoroughly cleaned, as needed, with the rubbing alcohol wipes.

- **Measurement of Cannabinoid Potency in Dry Samples**
  - The Luminary Profiler can be used to evaluate whole or ground dry cannabis samples. It is recommended, however, that the samples be ground to homogenize the plant matter and achieve more reproducible measurements.
  - After the sample window has been cleaned with the rubbing alcohol wipes, the dry plant material should be placed on the measurement window in such a way that the entire sample can be scanned.
window is covered. This will ensure that there are no regions where no sample is being evaluated by the NIR light source.

- After filling the measurement window with the sample, the black calibration cap is placed over the window to ensure that no stray light enters the system.
- The user then presses the Calculate button, and after approximately 10 seconds, the potency values for that specific aliquot of sample are provided.
- The user should remove the entire sample from the measurement window, and thoroughly clean the window with the rubbing alcohol wipes prior to measuring additional samples. Once the alcohol has evaporated, the next sample can be applied to the window.

### Measurement of Liquid Samples

- The Luminary™ Profiler comes equipped with an extract ring for the measurement of liquid samples, such as oils and concentrates. After following the calibration protocol, the user can select this type of sample.
- The extract ring must be thoroughly cleaned using the rubbing alcohol wipes. Prior to adding the sample, the ring must be measured to ensure that no NIR signal is produced from the ring and the external reflector. Place the ring on the measurement window, and then cap with the sample/black calibration cap, with the *silver* side facing down. Calibrate the instrument using this sample holder configuration.
- Add the liquid sample to the extract ring using a wide-orifice syringe. The user should add enough liquid to fill the sample well, and the sample should completely cover the measurement window. Failure to completely cover the measurement window may lead to analysis error. This procedure is predominantly for translucent samples. If a sample is dark or opaque, it may be necessary to add a thinner disc of sample to the measurement window, as highly darkened samples can prevent the reflected light from passing through the measurement window. In these instances, the measurement window should still be completely covered.
- After adding the sample to the measurement window, the external reflector (silver side of the black cap) is placed over the window. Since many of these types of samples are translucent, the NIR light will pass through the sample. In order to have the transmitted light reflect back through the measurement window, a reflective surface is needed. This process is termed “transfectance” since the light originally was transmitted through the sample, and then reflected back through the measurement window to be analyzed.
o As many of the liquid cannabis products are quite viscous, it is imperative to exhaustively clean any residual product from the measurement window and the extract between sample evaluations.

- Storage and Care of the Luminary™ Profiler
  o The Luminary™ Profiler should be stored in an area free of bright lights and strong airflow, and placed on a flat and stable work surface.
  o The daily calibration of the instrument will aid in ensuring maximum accuracy, and will normalize the system. The calibration should be performed when the instrument is turned on, and at least every 24 hours of continual use.
  o It is acceptable to leave the Luminary™ Profiler on when not in use; however, the Luminary™ Profiler is equipped with a lamp that has a finite lifetime. Therefore, when left on, and not in use, the lamp’s operational lifetime will be decreased. Replacement lamps are available from Sage Analytics.

b. Model Development for Accurate Predictions of Cannabinoid Potency

The Luminary™ Profiler combines traditional cannabinoid potency measurements from LC with NIR spectral data to develop multivariate analysis (MVA) models capable of accurately predicting THCa, THC, CBDa, CBD, and CBN contents. When only one analyte is measured, a standard univariate calibration curve is acceptable. Even if there are more than one species being quantified, a univariate calibration may work, if there are unique spectral features to each molecule. This is confounded when there are several analytes with similar chemical compositions, like cannabinoids. MVA enables to simultaneous quantification of all analytes, while significantly reducing the dimensionality of the data. This simply means that MVA reduces the amount of variables (each spectral point, for example) to hone in on those that are paramount to the model’s quantitative accuracy. Thus, several thousand wave numbers may be truncated to a few hundred or less, depending on the amount of information the data contains. The data generated from the LC analysis of the samples is united with the NIR spectra from the same strain aliquots.

The spectral data must be preprocessed to remove contributions from physical differences in the plants, such as particle size variation, color, etc., such that only chemical disparities influence the model’s development. Once the spectra have been effectively transformed, the model is produced using the reference data (LC) and the NIR transformed spectra. Ideally, the model should contain a diverse assortment of samples that encompass the expected analyte range of future samples. This is a key consideration, as the analyst must prognosticate what type of samples may be evaluated over time. To appropriately construct a model, the reference
samples should be split into randomly generated calibration and validation data sets. Initially, model accuracy can be gauged using a random or full cross-validation, depending on the total number of samples used for calibration. When a particular preprocessing method leads to suitable models (based on a statistical analysis of model performance), the model should be used to predict the cannabinoid content of the validation set. Since the potency values are known for this data set, the analyst can more accurately evaluate the robustness and validity of the model by assessing various metrics including the root mean standard error of cross-validation (RMSECV), the root mean standard error of prediction (RMSEP), the coefficient of correlation ($R^2$), the slope of the calibration line, the number of factors used to build the model (Scree plot), etc. The RMSEP and the number of factors are two of the most important parameters to monitor when building models. The RMSEP provides the error in the predicted values and also indicates the correct number of decimal places to include when reporting the potency value, as uncertainties should only be reported to one significant figure, since any further digits would be even less certain. If a model has a RMSEP value of 0.05%, the potency should be reported to two decimal places, 0.5%, one decimal place, and 5%, zero decimal places. Choosing an appropriate number of factors allows the most relevant spectral information to be included in the model, without over-fitting the data. When a model is over-fit, it attempts to explain random noise in the spectral data, a deleterious endeavor, as the accuracy of the model will plummet.

Another important metric to consider is the standard error of the laboratory, which is a gauge of the error in the reference method. This value is the lower limit of potential error in the model, as a model cannot out-perform the data used to generate it. Therefore, it is paramount to both know this value, and to recognize that a predictive model will never be more accurate than the reference method. The benefit of using NIR spectroscopy to develop models is that many more samples can be evaluated in time, compared to the reference method. This becomes an important point when considering that fact that very little of the bulk cannabis material is tested. In Colorado, for example, one gram out of every pound (454 grams) of cannabis is tested amounting to 0.2% of the product being deemed representative of the whole. The standard methods for analyzing this 0.2% are costly and time-consuming, limiting their utility for evaluating larger quantities of samples. Models constructed from reference and NIR spectral data enable the rapid, cost-effective, and accurate analysis of cannabinoid contents for all samples, providing a more realistic evaluation of the chemical constituents in cannabis strains as well as providing patients and recreational consumers a greater awareness as to what they are ingesting and inhaling.
The Quest for “True North” in Measuring Cannabis Potency
written by Jason S. Lupoi, Chad Lieber, Matt Kaplan, and Randall Kruep, Sage Analytics

As the medical cannabis industry has evolved, it has become apparent that there are various analytical methods to evaluate the cannabinoid content, or potency, of different strains. Knowing the cannabinoid content levels present in flowers, extracts/concentrates, and marijuana-infused products (MIPs) allows consumers to make educated decisions when purchasing products. However, laboratories use differing techniques such as high-performance liquid chromatography (HPLC) or gas chromatography (GC), leading to variable measurements of tetrahydrocannabinol (THC), cannabiol (CBN), and cannabidiol (CBD). In addition to the adoption of different instruments, analysts have varied experimental protocols within a defined analytical method. For example, in LC, technicians must select an appropriate column, eluent, flow rate, experimental run time, internal or external standard(s) to evaluate instrumental performance, and a suitable concentration range for measuring calibration standards, to name a few. The same experimental parameters must be selected in GC, as well as an appropriate carrier gas. Thus, the experimental protocols have become diverse, with different labs adopting different cannabinoid analysis strategies. This lack of standardization has led to the inability to actively compare cannabinoid metrics measured from different labs, necessitating a standardized protocol to be nominated and implemented henceforth. The adoption of a standardized method will allow scientists to contrast results between labs, and will enable round-robin style evaluations of the same sample to gauge the validity of the method, and potential lab-to-lab operator error. This method will also enable cannabis patients to select products in the same way they might choose an over-the-counter medication or other pharmaceutical products.

LC has been routinely used in industrial settings to separate and quantify analytes in a complex mixture. Samples must be in liquid form, necessitating the efficient extraction of compounds of interest from solid samples. A benefit of LC over GC is that the natural, acidic forms of THC and CBD can be quantified without requiring the derivitization of these molecules (as in GC) to enhance volatility. A downside of adopting LC over GC is the broader peak widths, resulting in decreased resolution of peaks with similar retention times. As mentioned above, there are various experimental parameters that must be optimized to obtain high peak resolution, such that samples with similar retention times can be differentiated and accurately quantified. These experimental conditions lead to some inherent shortcomings when seeking to measure a large volume of samples. For example, in the American Herbal Pharmacopeia, the run time chosen to provide the best separation of individual cannabinoid peaks was 30 minutes plus a six minute post-run to flush the column. If samples were measured using three representative aliquots per sample of interest, one sample would take 108 minutes. In addition, in order to obtain the isolated cannabinoid samples, the flower (bud) must be efficiently extracted. The extraction solvents employed (i.e. chloroform, methanol) are health hazards, requiring proper safety precautions to be adopted. Additionally, the eluent (acetonitrile and ammonium formate, for example, in the American Herbal Pharmacopeia) must be properly disposed. Thus, the
laborious sample preparation, long analysis times, and use of consumables limit the cost-effectiveness of using LC for analyzing thousands of samples.

GC techniques possess many of the same limitations when evaluating multitudes of samples. Additionally, unlike LC, GC cannot quantify the acid forms of THC and CBD, since the temperatures used decarboxylate the acid, forming neutral THC and CBD. Thus, the THC measurement provided is a gauge of the total THC contained in the plant (acid and neutral forms). GC is a destructive analytical technique, as the sample is converted to a gas prior to analysis. This feature translates to the loss of a valuable commodity, in order to obtain cannabinoid potency. The destruction of an aliquot of the sample could result in cannabis product vendors limiting how many samples they measure, leading to an erroneous representation of whole batches of sample by analyzing a small subset. The use of compressed gases presents a safety concern when using GC methods, and users must be extensively trained on how to safely use the gas cylinders, including how to change the regulators, how to properly store the cylinders, etc.

An underlying common theme when assessing the standard methods is that, although they are invaluable for analyzing smaller sample sets, their limitations prevent their use when higher throughput methods are desired to exhaustively evaluate cannabis samples. Vibrational spectroscopy has been routinely shown to relinquish the limitations of the standard methods when measuring plants (Lupoi et al., Bioenergy Research, 2014; Lupoi et al., Biotechnology for Biofuels, 2014; Lupoi et al., Bioenergy Research, 2015; Templeton et al., Cellulose, 2009; Shenk et al., Practical Spectroscopy, 2008; Ye et al., Bioresource Technology, 2008; Yamada et al., Holzforschung, 2006). Vibrational spectroscopy studies how light interacts with the sample of interest, and includes mid-infrared (MIR), near-infrared (NIR), and Raman spectroscopy. These methods are non-destructive, enabling users to retain their samples following the analysis. Another attribute of these techniques is the limited-to-no sample preparation requirement for obtaining the data. The high-throughput capabilities of these instrumental configurations enable researchers to thoroughly measure larger sample sets in less time and at decreased costs.

In order to take full advantage of the high-throughput characteristics, researchers often use spectroscopy in conjunction with one of the standard techniques to develop multivariate analysis models that are capable of predicting the analyte of interest accurately and robustly. For example, the Sage Analytics Luminary Profiler coupled GC data with NIR spectra to produce a partial least squares regression model for cannabis potency quantitation. These models are analogous to standard calibration curves, except they contain all of the important variables. Multivariate analysis enables the efficient mining of the spectral data to extract the useful information that may be obscured when visually analyzing the spectra. The following scenario can exemplify this process. If a lab has 2500 samples to evaluate, whether cannabis, aspirin, cereal grains, etc., the analysis via solely the standard methods would be a laborious and costly endeavor. The sample set can be divided such that the standard methods will be used to evaluate possibly 500-1000 of the samples. These samples are called calibration samples. NIR spectral data is then obtained for all 2500 samples, and the spectral data collected for the 500-1000
calibration samples is coupled to the standard results (i.e., GC or LC cannabis THC percent). After efficiently validating the models using a variety of statistical metrics, and ensuring the model is accurate, the remaining 1500 samples can be predicted. The root mean standard error of prediction (RMSEP) is the uncertainty to be applied in conjunction with the predicted values, and can be compared to the standard error of the laboratory (SEL), which is a metric indicative of the error in the standard method. Ideally, these two metrics should be close in magnitude, which translates to an accurate model that can be used confidently to quantify future unknown samples by merely acquiring the NIR spectrum, and inserting it into the model.

The cannabis industry should be subjected to the same rigorous testing procedures consumers have come to expect for any commodity. Would a consumer be comfortable purchasing a bulk supply of medicine, in which only one per every 1000 samples were analyzed? Even if 10, or 100 samples were deemed “representative of the whole”, can companies providing products expect consumers to blindly trust that a part really represents a whole? The Green Standard Working Group seeks to alleviate this conundrum by nominating a standardized testing method, such that large quantities of samples can be evaluated, giving a more realistic picture of cannabinoid potency across different strains. A standardized protocol would also enable labs to directly compare results to gauge natural sample variation, operator error, etc. Ideally, this method would incorporate an analytical tool, such as the Luminary Profiler, that can enable the whole cannabis eco-system (grow-houses, product manufacturers, dispensaries, and labs) to evaluate all samples that pass into their domain. The nomination of a standardized method will lead to a more proficient labeling of products, giving consumers detailed knowledge of the medicines and products they are purchasing, as they have come to expect and demand in as diverse of a portfolio of products such as peanut butter, sambuca, or aspirin.
Hardware Specifications of the Luminary Products

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<th>Profiler</th>
<th>Beacon</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dimensions (L x W x H)</td>
<td>230 x 340 x 160 mm</td>
<td>220 x 160 x 210 mm</td>
</tr>
<tr>
<td>Weight (lbs)</td>
<td>11.5</td>
<td>4.2</td>
</tr>
<tr>
<td>Power Consumption (W)</td>
<td>15</td>
<td>15</td>
</tr>
<tr>
<td>Input Voltage (VDC)</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>Lamp Type</td>
<td>Tungsten Halogen</td>
<td>Tungsten Halogen</td>
</tr>
<tr>
<td>Lamp Life (hrs)</td>
<td>2000</td>
<td>2000</td>
</tr>
<tr>
<td>User Interface</td>
<td>iPad Mini</td>
<td>On-Board Touchscreen</td>
</tr>
<tr>
<td>Environmental Rating</td>
<td>IP65</td>
<td>-</td>
</tr>
</tbody>
</table>

```
Spectral Range: Luminary Profilers and Beacons utilize the 1550-2000 nm window of the near-infrared fraction of the electromagnetic spectrum. This range was experimentally determined to be of the most value for the analysis of cannabinoids using analytical THC and CBD standards. Additionally, the measurement of THCA v. THC, and CBDA v. CBD standards was evaluated to add in determining unique peaks to acidic or neutral cannabinoids.
```

```
Number of Samples used to Develop Data Model:
Flower=399; Concentrates=557
```
Example of Lab v. Luminary Potency Comparison

FLOWER

CONCENTRATES
In the above plot, the x-axis shows the lab measured Total CBD (in concentrates) potency value, while the y-axis depicts that predicted from the Luminary. A table of the metrics associated with the above plot is provided below, and demonstrates the type of data that is used to evaluate whether a model is considered accurate and robust. The root mean standard error (RMSE) of prediction for an independent set of data is used to validate the model. This is the error one could expect in the predictions generated from the data model.

<table>
<thead>
<tr>
<th>Model</th>
<th>$R^2$</th>
<th>RMSE%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flower-THCA</td>
<td>0.89</td>
<td>3</td>
</tr>
<tr>
<td>Flower-CBDA</td>
<td>0.92</td>
<td>1</td>
</tr>
<tr>
<td>Flower-Total THC</td>
<td>0.90</td>
<td>3</td>
</tr>
<tr>
<td>Flower-Total CBD</td>
<td>0.94</td>
<td>1</td>
</tr>
<tr>
<td>Concentrates-THCA</td>
<td>0.91</td>
<td>8</td>
</tr>
<tr>
<td>Concentrates-THC</td>
<td>0.91</td>
<td>7</td>
</tr>
<tr>
<td>Concentrates-CBD</td>
<td>0.92</td>
<td>6</td>
</tr>
<tr>
<td>Concentrates-Total THC</td>
<td>0.95</td>
<td>6</td>
</tr>
<tr>
<td>Concentrates-Total CBD</td>
<td>0.95</td>
<td>4</td>
</tr>
<tr>
<td>Concentrates-CBN</td>
<td>0.16</td>
<td>1</td>
</tr>
</tbody>
</table>

- Example Metrics: Standard Error in the Luminary Predictions: +/- 10% relative standard deviation (RSD); this simply means that the error is ~10% of the potency value (20 +/- 2%, 40 +/- 4%, etc.). Although this number is rather ubiquitously given out by labs, we broke up regions of our plots into smaller ranges to get a better picture of what the error was in these specific regions.
Tips for Achieving the Best High-Throughput Experience with the Sage Luminary

Measuring Flowers

1. **Grind the sample** using the provided Sage Analytics grinder, or one of your own; due to the highly variable chemistry of any natural plant, homogenizing the sample is key to getting a realistic, accurate potency measurement.

2. Ensure the measurement window is clean, devoid of fingerprints, etc. prior to every measurement.

3. Fill the bud holder, and place the cap on top of the sample. You need to add enough ground cannabis to ensure that the bottom of the cap sits flush against the sample. Push down the cap to compress the sample as much as possible. The more contact between the sample and measurement window, the better. (Keep in mind that the instrument is not collecting light from the entire sample. Rather, if you look at the light source striking the window, you will see some glare in the center of the measurement window. This is where the instrument is collecting light. Also, the light penetrates about 1mm into the sample; the light is not penetrating through the entire sample.)

Measuring Concentrates

The disposable sample cell provides users of the Sage Luminary products with a simple sample preparation to evaluate concentrate samples. While the overall process is fairly easy, improper use of the sample cell can lead to inaccurate results.

1. Before adding a sample to the disposable, **be sure to obtain the blank measurement of the empty cell**, as defined on your iPad or touchscreen software interface. Measuring a different sample cell can lead to inaccuracies in your data. If you pre-fill all of the disposables with your samples, you will have to measure a different disposable as a blank. Although these disposables are crafted to be as uniformly similar as possible, there are going to be subtle variations in thickness and perhaps in the molding of the cell. **Remember: measure the blank, then add the sample, and measure.**

2. When adding concentrate to the sample well, **ensure that the entire well is filled**, prior to placing the disposable on the instrument for analysis. **Double-check that the entire well is filled, and that there are no bubbles in the sample.** Bubbles will significantly alter the potency results. Sometimes, the liquid sample can migrate to one side of the cell, producing a half-moon effect of sample. Should this happen, **press down on the silver side of the disposable.** If pressing down still doesn't cause the sample to be uniform throughout the disposable well, add more concentrate to the well. Repeat the process as needed until there are no bubbles or empty regions in the sample well.

3. When closing the disposable sample cell, you should handle by the edges, and snap shut. The bottom of the disposable should not be touched with bare hands, as fingerprints and oils from your skin can adhere to the plastic, and foul the measurements. If you did handle the bottom of the cell, gently wipe with an alcohol wipe, and allow to dry.

4. When measuring dark samples like shatter, **use just enough to fill the sample well.** Overfilling with these types of materials can result in potency differences. Dark samples tend to absorb light. The more light that is absorbed by a sample, the less light that is reflected through the instrument. Less light equals lower potencies. If you can see a bulge in the disposable, looking down at the silver side, you should remove a little of the sample. Keep in mind that the shatter/wax should be compressed by pressing down on the silver side after filling the cell, and snapping shut.
### Flower Sample Prep

- **Under Filled Bud Holder**
  - (Light is shining through)

- **Perfectly Filled Bud Holder**
  - (No light is shining through)

### Concentrate Sample Prep

- **Under Filled CO2 Oil**
  - (bubbles are present)

- **Perfectly Filled CO2 Oil**
  - (no bubbles present)

### Other Sample Prep

- **Under Filled Shatter**
- **Perfectly Filled Shatter**
- **Over Filled Shatter**
Step 1: Ensure the measurement window is clean using the alcohol wipes.  
*Note: Make sure the window is completely dry before proceeding.*

Step 2: Dark Calibration:  
Place the black cap over the measurement window and select NEXT.

Step 3: White Calibration:  
Place the white reference puck over the measurement window and select NEXT.  
*Note: Please store puck right away, when done.*
Step 4: Grind the cured sample to reduct particle sizes and achieve better homogeneity. Alternatively, the samples can be sieved using a 1.4mm mesh size (No. 14 sieve).

Step 5: Fill the bud holder with the ground bud. 300mg should suffice. Note: No gaps or light should be seen through the sample.

Step 6: Use the cap to compress the sample. Once compressed, add a small amount more and compress again. Using about 300 mg should be sufficient.
Step 1: Ensure the measurement window is clean using the alcohol wipes.  
*Note: Make sure the window is completely dry before proceeding.*

Step 2: Dark Calibration:  
Place the black cap over the measurement window and select NEXT.

Step 3: White Calibration:  
Place the white reference puck over the measurement window and select NEXT.  
*Note: Please store puck right away, when done.*
Step 4: Measure the empty cell by placing on the window, and selecting NEXT. This is really important, as it removes any potential signal from the cell itself. *Failure to do this can lead to incorrect potency values.*

Step 5: Add the oil to the empty cell. For the best measurements, apply just enough concentrate to fill the well (~100 μL). Check for bubbles, and to ensure the well is completely covered. Snap shut, being careful to NOT touch the bottom, where the sample well is located. Fingerprints can cause error in the measurements.

Step 5a: Sample well expanded view. *Note: You only need to add to the sample well itself, not the entire inside of the cell.*

- Improperly filled disposable sample cell
- Problem: Not uniform distribution in sample well
- Properly filled disposable sample cell
- A good, even distribution has been reached
<table>
<thead>
<tr>
<th><strong>Lab Analysis</strong></th>
<th><strong>Sage Analytics Analysis</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>GC or HPLC</strong></td>
<td><strong>Luminary Beacon</strong></td>
</tr>
</tbody>
</table>

1. The tester or quality control agent pulls a small sample from the bag.
2. Entire testing is done by a single person within a 10 second time frame, so no ID numbers, data entry or need to worry about “chain of custody”.
3. The dried bud is ground into a homogeneous substance or the extract is placed directly in the sample cell.
4. No solvents or harmful chemicals needed - simply uses light.
5. Analysis by a $23,900 (new) Luminary Beacon that can determine Total THC, THCA, Total CBD, CBDA and CBN using optical spectroscopy.
6. Can be run by anyone with minimal training.
7. Samples are not discarded and remain completely intact allowing it to be reused for another purpose (i.e. taste testing, turned into a pre-roll or edible).
8. A CannaMetric Profile label immediately prints out showing the potency measurement results and can be applied directly to product container.

**Total time from start to finish:** 2-3 days  
**Total Cost:** $100 per test  
**Tests per day:** 24 in an 8 hour period

**Total time from start to finish:** 20 seconds  
**Total Cost:** Pennies per test  
**Tests per day:** 200 in an 8 hour period
**GAS Chromatograph**

1. Heated Injection Port
2. Column
3. Flame Ionization Detector
4. THC, CBD Results

**LIQUID Chromatograph**

1. Pump
2. Column
3. UV Detector
4. THC-acid, CBD-acid Results

**LUMINARY BEACON**

1. Dry Bud or Extract (no solvent required)
2. Place ground bud on measurement window or extract in disposable sample cell
3. Close lid and press “Calculate”
4. View results and print out “Cannametric Profile Label”

**TOTAL TESTING TIME**

- Gas Chromatograph: 20 MINUTES
- Liquid Chromatograph: 20 SECONDS

**Total THC, THC-acid, Total CBD, CBD-acid, CBN Results**

www.sageanalytics.com
Sage Analytics Common Questions & Answers

1. **My Luminary Profiler™ / Beacon™ won’t turn on**
   - Is it plugged into a power source?
   - Is the plug damaged?
   - Has the power button been activated?

   If all these things have been done and the device still doesn’t turn on, the internal lightbulb may be damaged - you need to call for a replacement bulb.

2. **Why do I need to wait 20 minutes for the system to warm up?**
   The system needs time for the optical components to reach a steady temperature, so it can produce the most stable results. This is a crucial step to obtaining accurate potency data.

3. **Why do I need to calibrate the device every time I turn it on?**
   The white and black calibrations (akin to a light and dark balance on a camera) are necessary to account for any slight variations in the lightbulb from day to day. These calibrations are critical to the measurements, so special care should be used to make absolutely sure both the Luminary window and the white calibration puck are completely clean, that the white puck is properly centered on the Luminary window, and that no vibrations or jarring of the instrument occurs during the calibration procedure.

4. **I've tested the same sample three times in a row and I got slightly different readings each time. Why?**
   The Luminary measures an optical signal then converts this into a potency value. The light that reflects off the sample has some randomness to it, which causes very slight differences that may result in different potency readings. Furthermore, there is a margin of error in the predicted value, which is on the order of 10% of the measured value. That means, for example, that a sample with 20% THC would produce a reading between 18-22% (10% of the 20% reading is 2%, so 20% +/- 2%).

5. **Can I measure whole buds or do I need to grind the bud?**
   Since the cannabinoid density varies considerably across buds, it is always recommended to grind the bud for a more representative reading of the sample.

5a. **I measured a whole bud and got one reading and then I turned it over and measured it again and got a completely different reading...why?**
   Cannabis is a natural product, and nature has a lot of variety, as per the description to #5, above. Additionally, if the sample moved between measurements (from bumps on the table, vibrations, or a breeze) then the readings will be different since it would be measuring a different region of the bud.
6. The label printer isn’t working.
   • Is the white cord plugged into the back of the Luminary or Beacon?
   • Is the printer itself plugged into the wall?

6a. I can hear the label printer working, but nothing is printing on the labels.
Try flipping the labels over. These are labels specific for our thermal label printer, and therefore, if the labels are not properly loaded, you will not see anything printed on the label backing.

7. Why is my potency testing result different than what the Lab I use indicates?
The lab tested a bud or a couple of buds, but not the bud you just tested. Natural variation causes a huge difference in potency between different plants, between different buds, the same plant and even within the same bud. On top of this natural variation, there is very little standardization in how different labs produce their result. The labs use different measurement technologies, different techniques to obtain the measurement, and have their own margin of error in their test result. Combining these two factors (natural variation and lack of laboratory standardization) means that it is much more likely that your result will differ.

8. What is the standard error for the Luminary?
The error on each reported cannabinoid value is on the order of 10% of the measured value. That means, for example, that a sample with 20% THC would produce a reading between 18-22% (10% of the 20% reading is 2%, so 20% +/- 2%).

9. What is the difference between THCA and Total THC? What is the difference between CBDA and Total CBD?
The A in these cannabinoids refers to their acid form, and is the form of the cannabinoids that is most prevalent in the plants. Converting the acids to their neutral forms is done by heating (called decarboxylation), and it is the neutral form of THC that has psychoactive properties. The total THC is the sum of the neutral and the acid forms; same thing for CBDA and Total CBD. This summation does not use the formula for determining total potential Δ9-THC.

10. How much of a sample should I use to obtain the most accurate reading?
For bud, ground product should loosely fill the bud holder, coming all the way to the top, then compressed by fully inserting the cap. Overfilling isn’t a problem, but under filling will lead to errors. For extracts that use the disposable sample holders, the round dimple in the center of the sample holder must be completely full, leaving no space around this dimple, and devoid of bubbles or other gaps that would allow light to pass through without hitting the sample. The sample holder should not be overfilled, causing the sample holder to bulge. If you notice a bubble, or an uneven distribution of the sample in the dimple, flip the cell over on your counter, and press down on the silver circle. This may correct the sample filling issues. If not, add more sample until you achieve an even distribution, or the bubble is removed.

11. What type of maintenance do I need to do on the Luminary / Beacon?
Keeping the measurement window clean is imperative, and is best accomplished using an alcohol wipe followed by a paper towel or tissue (to remove the haze from the alcohol wipe). Same goes for the white reference puck. Beyond those, changing the lightbulb when it burns out is the only real maintenance.
12. I spilled water/soda/beer on my device and now it won’t turn on. What should I do? Is it still covered under warranty?
Wipe up as much of the liquid as possible. If you have a Luminary Profiler, you can wipe it down with a wet rag and it should still work just fine. If you have a Luminary Beacon, let the device sit in a well-ventilated area for at least a day to allow any liquid inside to dry out. If it still doesn’t turn on, allow the system a bit more time for any liquid to evaporate. If it still won’t power on, call Sage technical support for return options. While damage like this is not covered under warranty, it still may be repairable.

13. How do I clean the device?
The window should be cleaned with an alcohol wipe followed by a dry, lint-free cloth. The screen and the device can be wiped with a dry cloth. Solvents and cleaners may damage the screen or the system finish.

14. Can I leave the device on overnight?
Sure you can. It will require a new calibration after 24 hours, however.

16. I put something other than cannabis on the machine just to see what it would do and it gave me a potency measurement reading.
The Luminary is a cannabis potency profiler, not a cannabis detector. Many plants and herbs have chemical profiles very similar to cannabis, and may generate false potency readings.

17. Do I need to put it back in its storage case or can I leave it out on the counter for extended periods of time.
That’s up to you.

18. How do temperature extremes affect the readings?
Wide temperature swings can cause higher or lower potency readings. It is recommended that the machine be kept in a moderate room temperature (~60-85F), and that the readings be performed at the same general temperature as when the device was last calibrated. If the room temperature changes enough that you can feel the change, a recalibration would be advised.

19. I lost my white reference puck...how do I order replacement parts?
Call Sage sales.

20. How long will the light bulb last and how do I order a new one?
The bulb is rated for about 2,000 hours of usage. Call Sage sales to order a new one, and consider picking up a spare so you can minimize downtime.

21. Is the Luminary / Beacon a state certified device? Can I just use this and not send out to the Lab anymore?
The Luminary and the Beacon were “trained” utilizing samples from labs that were either state or otherwise certified. Each state has their own certification and we have not sought approval from each individual state. If your state requires you to test, you must still send to a state certified lab.
22. The touchscreen is frozen and won’t work.
First try restarting the unit. If that doesn’t fix it, make sure the machine is plugged into an outlet that doesn’t share connections with multiple other devices. “Dirty” electrical power from the wall is a frequent culprit to touchscreen issues. If these don’t fix the sensitivity issues, call Sage technical support for return/repair options.

23. The machine feels extremely hot to the touch…should I be worried?
It is normal for the machine to get warm, but it should not get hot. First make sure that the machine is not being used in a high temperature environment (~85F and up). Try powering down the system and allowing it to cool off before restarting. If it continues to get hot, call Sage technical support for repair options.

24. If I test a sample, can I ingest it afterwards?
The machine doesn’t alter the sample in any way, so what you do with it after you measure it is your decision.

25. I ran out of disposable sample cells, how do I order more?
Call Sage sales.

26. Do I have to use the disposable sample cell for extracts or can I just place it directly on the measurement window? Will I get a different reading?
Yes, you must use the disposable sample holder. Placing samples other than bud on the window will lead to erroneous measurements.

27. Can I use the disposable sample cell for dry bud as well?
No, the dry bud must be placed in the bud holder. The disposable sample cell is for all products other than bud.

28. What is the best way to clean the measurement window?
Wipe the window with an alcohol wipe or a paper towel and ethanol. Then wipe with a dry, lint-free tissue or paper towel to remove any haze that may be present from the alcohol wipe.

29. Why do I need to clean the measurement window every time I test a new sample?
Oils from the sample can collect on the measurement window, which can foul the instrument’s accuracy. Also, cleaning the window will eliminate the chances that fingerprints, dust, etc. are contaminating the measurement window.

30. Do I need to have Internet access in order to test samples?
No. Future developments will add further functionality via web connection, but the potency measurement does not require Internet connectivity.