Analysis of elemental contaminants in cannabis and hemp using inductively coupled plasma mass spectrometry (ICP-MS)

Complete **CANNABIS TESTING SOLUTIONS** for the Canadian market

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Goal
To show the accurate and precise analysis of elemental contaminants at low levels in relevant sample matrices.

Introduction
With recent shifts in legislation regarding the production and consumption of cannabis and related products, regulations are starting to emerge regarding potency, impurities, and contaminants. One major set of contaminants that require testing are heavy metals due to their toxicity and/or carcinogenicity.

Metals are known to accumulate in plant material through normal metabolic processes and some plants (including cannabis and hemp) are capable of hyperaccumulating metals. Hyperaccumulation in plants can lead to drastically increased metal concentrations and potential toxicity risks. For Canada, guidance for the minimum limit and testing requirements for heavy metals (arsenic, cadmium, mercury, and lead) in cannabis and related products are based on USP chapters <232> and <233>, with the limits based on those for the inhalation of drug products. The applicable limits for the aforementioned elements are shown in Table 1.

Table 1. Applicable limits for the big four for inhaled drug products

Element	Limit [$\mu g \cdot g^{-1}$]
Arsenic (As)	0.2
Cadmium (Cd)	0.5
Mercury (Hg)	0.2
Lead (Pb)	0.1

To maintain regulatory compliance, it is important that cannabis cultivators have access to a simple, robust, multi-elemental analytical tool that provides accurate and precise concentrations of toxic metals in cannabis and related products. This includes sample preparation for different sample matrices, such as flowers, edibles or oils. The required low limits of detection (considering added dilution factors through sample digestion) make inductively coupled plasma mass spectrometry (ICP-MS) a perfect technique for this application. In addition, ICP-MS offers multielement analysis capacity (more than 60 elements can be easily analyzed in one aspiration of a sample), and its wide dynamic range make it uniquely positioned for this type of analysis. At the same time, ICP-MS can be combined with dedicated sampling accessories like autodilutors, which can help to increase laboratory productivity and reduce the risk of error through human preparation.

In this study, a method for the accurate determination of heavy metals in cannabis is outlined, including microwave assisted digestion for sample preparation and automated in-line dilution for automated preparation of calibration standards and sample dilution. The combination of ICP-MS with an automated dilution system improves laboratory throughput and reduces human interaction with the samples, liberating operator time for other tasks and avoiding errors in the process of sample preparation and dilution.

Method

Sample preparation

In total, 10 different cannabis samples and 5 hemp samples were prepared for analysis. To approximately 0.25–0.5 g of each (previously homogenized) sample, 10 mL of HNO_3 and 2 mL HCl were added in a closed PTFE vessel. After the initial reaction had ended, the samples were digested within a microwave (NovaWave, SCP Science, Quebec, Canada) according to the program provided in Table 2.

Table 2. Time and temperature settings for microwave digestion

Mode	Temperature (˚C)	Time (min)
Ramp	200	15
Hold	200	45
Cooldown	200	30

After complete cooling of the digested samples to ambient temperature, 2 mL of digested sample were transferred into 50 mL polypropylene tubes followed by addition of 48 mL of ultrapure water (ASTM Type 1 with a resistivity of >18.2 MΩ·cm). Any further dilutions were accomplished using an autodilution system. All prepared solutions for matrix, dilution, and carrier were prepared using 5% HNO_3 (v/v), 1% HCl (v/v). All calibration and quality control solutions were prepared in the same aqueous media as the samples. To all solutions, 100 mg·kg^{-1} of gold was added to reduce potential memory effects of mercury. The acids used for digestion and preparation of standard solutions were of trace metal grade (Fisher Scientific). All standard solutions were prepared gravimetrically using ultrapure water.

Instrument configuration

The instrument used for analysis was a Thermo Scientific™ iCAP™ RQ ICP-MS in conjunction with a prep*FAST*™ SC-4DX autosampler and autodilution system (Elemental Scientific, Omaha, NE). The combination of these two systems allows for a completely integrated sampling and analysis station, with the option to automatically and intelligently dilute all samples and standard solutions. The instrument´s performance was evaluated on each day it was used. To access the instrument´s performance (sensitivity as well as key indicators, such as oxides and doubly charged ratio), a performance report was run using the Thermo Scientific™ Qtegra™ Intelligent Scientific Data Solution™ (ISDS) Software. In case a deviation from the required performance specifications was detected, an autotune routine was conducted automatically. The typical operating conditions are summarized in Table 3.

Table 3. Typical operating conditions for the iCAP RQ ICP-MS

Parameter	Value
Spraychamber	Quartz baffled cyclonic
Nebulizer	PFA-ST
Injector	2.5 mm i.d. quartz
Interface	Nickel sample and skimmer cone, High Matrix insert
Forward power	1550 W
Nebulizer gas	1.0 L·min^{-1}
QCell Settings	4.5 mL min^{-1} He, 3V KED Voltage
Sample uptake/wash time	10 s each
Dwell times	Optimized per analyte, 50–200 ms per peak, 10 sweeps
Total acquisition time per sample	2:45 min

To remove all interferences potentially causing a false positive bias in the results, such as $^{40}Ar^{35}Cl^+$, interfering on $^{75}As^+$, the instrument was operated using kinetic energy discrimination (KED). For all unknown samples, a full mass spectrum was acquired to detect all elements present in the sample. This way, the presence of other contaminants can be qualitatively and semi-quantitatively determined later.

Addition of internal standards (^{45}Sc, ^{89}Y, ^{115}In, ^{159}Tb, and ^{209}Bi; at a concentration of 5 ng·mL^{-1} each) for correction of potentially occurring matrix effects in the samples was done automatically using the prep*FAST* autodilution system. All data handling and calculations were performed using the Qtegra ISDS Software, including the generation of calibration curves, determination of the Instrumental Detection Limits (IDLs) and calculation of recovery limits for quality control samples such as the Initial Calibration Verification (ICV), Continuing Calibration Verification (CCV) and the matrix spike.

Results and discussion

After calibration of the instrument, all samples were analyzed to determine the concentrations of the respective impurities. All samples contained concentrations of the analytes under study well below the required limits in Canada. Instrumental detection limits were determined based on the standard deviation of the signal in a blank sample (only containing the acid matrix and gold), but as well for a sample spiked with relevant quantities of all analytes. In all cases, IDLs were found to be in the single digit ng·L^{-1} range (between 2 and 7 ng·L^{-1}). This in turn leads to method detection limits (MDLs) below 1 ng·g^{-1}, considering all steps for sample preparation.

To assess the effect of the sample preparation, samples were spiked with 500 ng·g^{-1} As, Pb, Cd, and Hg and the spike recoveries were calculated. In all samples, the spike recovery was found to be within acceptable limits (70–150% as per USP chapter <233>). All data is summarized in Table 4.

Table 4. Results of the analysis of hemp and cannabis samples and spike recovery tests

Sample	Concentration unspiked [ng·g^{-1}]				Spike recovery [%]			
	^{75}As	^{111}Cd	^{202}Hg	^{208}Pb	^{75}As	^{111}Cd	^{202}Hg	^{208}Pb
Hemp 1	0.13	0.26	0.04	0.65	104.7	103.9	119.9	107.5
Hemp 2	0.22	0.78	0.06	0.71	101.7	100.9	118.7	108.7
Hemp 3	0.01	n.d.	n.d.	0.05	114.4	103.4	101.6	94.9
Hemp 4	0.70	1.25	n.d.	1.35	97.6	99.8	108.5	102.0
Hemp 5	1.22	0.09	n.d.	1.1	88.8	106.9	106.8	106.1
Cannabis 1	n.d.	n.d.	n.d.	0.02	122.9	116.4	115.7	103.9
Cannabis 2	n.d.	n.d.	n.d.	0.03	117.7	114.6	113.1	103.7
Cannabis 3	n.d.	n.d.	n.d.	0.04	126.8	116.5	114.7	101.3
Cannabis 4	n.d.	n.d.	n.d.	0.03	122.9	113.9	113.6	103.4
Cannabis 5	n.d.	n.d.	n.d.	0.02	116.7	111.2	110.5	104.0
Cannabis 6	n.d.	n.d.	n.d.	0.03	124.1	114.6	113.3	102.3
Cannabis 7	0.07	0.01	n.d.	0.05	99.6	110.5	122.7	109.1
Cannabis 8	0.09	n.d.	n.d.	0.09	96.7	108.1	120.0	107.4
Cannabis 9	0.07	n.d.	n.d.	0.06	96.6	107.1	118.6	109.0
Cannabis 10	n.d.	n.d.	n.d.	0.03	128.2	116.1	112.4	102.0

As can be seen from the results, hemp samples contained slightly higher amounts of the impurities under investigation in comparison to cannabis samples. Also, the obtained results for spike recovery indicate that the proposed method can detect trace amounts of the regulated toxic elements with good accuracy. In most cases, excellent spike recoveries between 90% and 110% are obtained. In some cases, however, slightly elevated spike recoveries are observed in samples where the impurities were found to not be present in the unspiked (for example, for ^{75}As in cannabis samples 1–6 and 10). As the amount of sample used for digestion was variable between 0.25 g and 0.5 g, it cannot be excluded that this effect could be caused by residual carbon from the digestion process, leading to a slight increase in the responses for arsenic, mercury and cadmium through carbon enhancement. The potential presence of rare earth elements, such as neodymium or samarium, potentially creating $[M]^{++}$ interferences unresolvable using KED, could be excluded based on the examination of full mass spectra.

Quality control

During the analysis sequence, comprising both sample types with unspiked and spiked samples, applicable quality control tests were conducted after the analysis of 10 samples. In total, more than 30 unknown samples could be run in 3.5 hours, including all blanks, calibration standards, and quality control samples (ICV, CCV, and spike recovery tests). The results for ICV and CCV tests are displayed in Table 5.

Table 5. Initial and continuing calibration verification results from a 3.5 hour run

Sample	^{75}As	^{111}Cd	^{202}Hg	^{208}Pb
ICV	104.0	101.5	90.7	102.5
CCV 1	103.0	101.1	86.9	102.4
CCV 2	113.6	102.4	89.4	102.7
CCV 3	109.2	102.6	87.3	101.9
CCV 4	106.2	102.2	85.4	102.4

Figure 1 shows the recovery of the internal standards monitored over the entire runtime of the batch. All internal standards are found to be well within the regulatory limits. Samples 46 and 48, and 53 and 55, corresponding to two specific hemp samples (each spiked and unspiked), showed significantly increased response of the internal standard Y. This is likely caused by an increased amount of strontium present in the samples, and hence formation of $^{88}Sr^{1}H^{+}$, either by natural variation or a larger portion of sample used for preparation. Based on the setting for intelligent automatic inline dilution, all samples were automatically reanalyzed, applying an additional 10-fold dilution. The internal standard response of the automatically reanalyzed samples can also be seen in Figure 1, directly following the original sample (i.e., at position 47, 49, 54, and 56). In both cases, the internal standard response reached acceptable limits and correct results were obtained for spike recovery. Additionally, for both of these hemp samples, examination of a full spectrum (acquired for each unknown sample) revealed a drastically higher concentration of various elements, including potassium, iron, or barium, potentially leading to the observed effect.
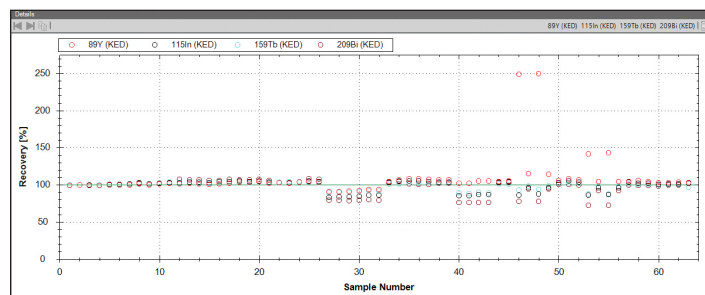

Figure 1. Internal standard response over the entire runtime of the analysis. Generally, response is well within the acceptable range, but two samples (46 and 53 with their spiked equivalents 48 and 55, respectively) show a different behavior.

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Conclusion

The iCAP RQ ICP-MS can accurately and reliably quantify elemental contaminants in hemp and related products with high accuracy and precision. Polyatomic interferences, affecting, for example, the detection of arsenic or cadmium are effectively removed using KED. With the unique design of the QCell collision/reaction cell system, it is possible to analyze all elements, regardless of lower mass, such as ^{9}Be, or higher mass, such as ^{208}Pb, using the same conditions, effectively reducing the required analysis time per sample and contributing to laboratory productivity. Automatic dilution is an effective way to further enhance the laboratory throughput and to better leverage the most critical resource in a lab, the time available for laboratory technicians.

Acknowledgement

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