



THOMPSON RIVERS UNIVERSITY

Directed Studies 2026 Winter Report

**Application of Microwave-Assisted Acid Digestion and ICP-MS
for Determination of Major and Trace Elements in Bee Pollens**

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of plants and consequently being incorporated into plant materials such as pollen.⁸ However, it should be noted that although heavy metals may be in a plant's immediate environment in the soil, it does not mean that the plant will uptake all the metals present and incorporate them into its materials. Not only do plants have mechanisms to selectively uptake certain metals while avoiding others for their own health, the solubility and interactions of each metal with the soil particulates also determine their uptake.⁹

As bees collect pollen from these potentially metal polluted plants, contaminants may be incorporated into bee pollen, creating health concerns for both the bees and humans who consume it. The presence of heavy metals, even at low concentrations, are toxic and can result in adverse health effects such as kidney dysfunction (Cd), reproductive toxicity (Pb), cancer (As), and respiratory failure (Hg).⁸ In addition, heavy metal environmental contamination reduces bee species diversity and richness; however, abundance remains stable.¹⁰ This is highly significant as bees pollinate most of the planet's ecosystems and without their contribution, it would likely collapse.¹

Given these concerns, accurate quantification of elemental concentrations in bee pollen is essential. Inductively coupled plasma-mass spectrometry (ICP-MS) is a highly sensitive and accurate technique capable of detecting very small elemental concentrations (ppb). When combined with microwave-assisted acid digestion for the preparation of samples, ICP-MS appears to be a reliable and accurate approach for the determination of trace and heavy metal concentrations in bee pollen.¹¹ The performance of this method can then be evaluated through assessment of detection limits and precision.

Methods

Sample Collection and Preparation

Bee pollen samples were obtained from five locations in Canada along with one location in Spain. Table 1 illustrates each bee pollen sample's brand name, location, and abbreviation used for this report. All bee pollen samples were stored at room temperature away from sun. Prior to digestion, around 0.5 g of bee pollen was ground into a fine powder via mortar and pestle to ensure homogeneity.

Table 1. Bee Pollen Samples

Abbreviation	Brand Name	Location
DGO	Dutchman's Gold Pure Bee Pollen	Carlisle, ON
PBS	Plant Bee Spain	Spain
RMB	Rocky Mountain Honey Farm	Castledale, BC
PBC	Planet Bee Saskatoon	Saskatoon, Saskatchewan
DBA	Dr. Bee Pollen	Pitt Meadows, BC
MVE	Meadow Vista Bee Pollen	Kelowna, BC

Microwave-Assisted Acid Digestion

Approximately 0.5 g of each sample was then transferred to an Anton Paar acid digestion vessel, followed by the addition of about 10 mL of concentrated nitric acid. Digestion vessels were sealed and digested using an Anton Paar Multiwave Go Plus microwave digestion system with a 10-minute temperature ramp to 150 °C that was held for 15 minutes, followed by a 15-minute cooldown to room temperature. After digestion, solutions were diluted with approximately 15 mL of 2% nitric acid (or 2% nitric acid + 0.5% hydrochloric acid for mercury specific analysis). The solutions were then transferred to a 50 mL Falcon tube through filter paper in a filter funnel before being diluted to a total volume of 50.00 mL with the same solution used previously for dilution.

Standard Preparation

Calibration standards were prepared in six concentration levels from 200 ppb intermediate stock solutions. To prepare the 200 ppb intermediate stocks, 1 mL of the original 10 ppm stock was diluted to 50 mL with 18 MΩ water. Three environmental standard mixes were used in this study: one including Ag, Al, As, Ba, Be, Cd, Co, Cr, Cu, Mn, Mo, Ni, Pb, Sb, Se, Th, Tl, U, V, Zn; another including Ca, Fe, K, Mg, Na; and the last with only Hg. Standards were diluted with 2% nitric acid for standard preparation, except for the Hg standards, which were diluted with 2% nitric acid + 0.5% hydrochloric acid.

Table 2. Concentrations of Standard Levels Used

Standard Level	Volume 200 ppb Environmental Stock (mL)	Total Volume (mL)	Concentration (ppb)
0	0.000	50.00	0.000
1	0.025	50.00	0.100
2	0.250	50.00	1.000
3	2.500	50.00	10.00
4	12.50	50.00	50.00
5	25.00	50.00	100.0

ICP-MS Analysis

Elemental analysis was performed using an Agilent 7900 ICP-MS. Argon plasma was used with helium and hydrogen as collision gases to reduce polyatomic interferences.

Method Validation

Method performance was evaluated in terms of limits of detection (LOD), limits of quantitation (LOQ), and precision. Intraday precision was determined from triplicate measurements from each run, while interday precision was approximated from the total pool of sample measurements over intraday runs. Precision was expressed as percent relative standard deviation (%RSD).

Results

Method Performance

Calibration curves showed strong linearity with R^2 values above 0.99 for most elements (Tables 3-4). The exceptions to this observation were elements often present at larger concentrations such as Na, Mg, K, Ca, and Fe ($R^2 > 0.98$) along with Hg ($R^2 > 0.88$) which often has a minute presence in samples. Instrumental limits of detection (LOD) were also provided for each elemental analysis. Calculated average LODs and limits of quantitation (LOQ) demonstrated extreme instrumental sensitivity as elemental LOQs were near or below 1 ppb. Intraday precisions were calculated for each element as an average among all analyzed samples (Table 5). Additionally, interday precision was determined from all measured values across runs. Some elements displayed acceptable precision with values less than 15% RSD; however, most intraday