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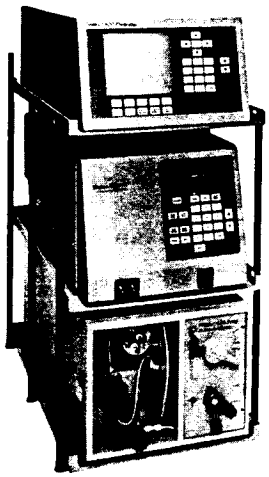
"...an analytical lab incorporating microwave digestion followed by CIA can process...200 cation analyses per day."

New U.S. laws for nutritional labeling of foods mandate the labeling of sodium and calcium and make the labeling of potassium voluntary. Obviously, this has led to an increased demand for sample analysis of these cations. Currently, the analysis of cationic nutrients in foods is a routine procedure using atomic absorption spectroscopy (AAS) or inductively-coupled plasma spectroscopy (ICP). The former is a tedious process allowing only single analyte determination; the latter represents a significant investment in instrumentation. In both cases, microwave or hot-plate acid digestion as well as combustion oven ashing are used for sample preparation. Overall, these techniques are lengthy and require time consuming, hands-on sample preparation. Ion chromatography (IC) and capillary ion analysis (CIA) may provide more efficient, higher throughput methods for the analysis of these cations.

Ion Chromatography and Capillary Ion Analysis: Greater Efficiency and Higher Throughput

Ion chromatography chemistries, such as the Waters IC-Pak™ C M/D column, separate monovalent and divalent cations in a single analysis. The IC-Pak C M/D column is based on "coordination ion chromatography" which is performed on a poly (butadiene-malic acid) -coated silica column with an EDTA-nitric acid isocratic eluent. This chemistry, coupled with conductimetric detection, allows you to measure multiple nutritionally relevant cations such as sodium, potassium, magnesium and calcium in a single assay.[1,2]

Capillary ion analysis is a relatively new technique which holds great promise



The Waters Action™ Analyzer and IC-Pak™ C M/D column separate monovalent and divalent cations in a single analysis.

for the analysis of cations from foods. A preliminary evaluation of the application of CIA is discussed here. All sample digests were introduced into a capillary electrophoresis system for capillary ion analysis.[4] Good separations of the analytes of interest with no sample induced baseline upsets were observed with this technology. The results were compared to results obtained by AA.

Microwave Digestion for Rapid Sample Preparation

Microwave technology has made a broad entry into the analytical lab for a variety of sample preparation procedures. Microwave acid digestion of food matrices has been shown to be a fast way to prepare samples prior to cationic nutrient determination by AA.[3] Ion chromatography and CIA can simultaneously quantify multiple analytes and can easily be automated. For laboratories looking for a way to maximize throughput for cation analysis, IC or CIA and microwave digestion is an ideal combination.

This study demonstrates the feasibility of the combined approaches of CIA/microwave digestion and IC/microwave digestion. The microwave digestion procedure was optimized—relative to sample size and which acids to use—in order to provide a digest that was compatible with the chromatographic and electrophoretic processes. Sample concentrations and dilutions were optimized relative to the sensitivity/mass load capabilities of the chromatography and electrophoresis. A variety of food samples were chosen to represent diverse matrices that could potentially challenge a food analysis method.

Experimental

Instrumentation. The instrument used for the ion chromatography portion of the study was a Waters Action Analyzer™ ion chromatograph equipped with a Model 431 conductivity detector and a Waters 717plus autoinjector. A Millennium® 2010 Chromatography Manager was used to collect, integrate and store data.

For the CIA feasibility study, a Waters Capillary Ion Analyzer with CIA chemistry was used also with the Millennium 2010 Chromatography Manager.

Eluents and Electrolytes. The eluent used for the ion chromatography was a solution of 0.1mM ethylenediaminetetraacetic acid (EDTA free acid) and 3.0mM nitric acid.

The electrolyte used for the CIA separations was a 1.2mM UV-Cat-2™ (available from Waters Corporation) and 3.0mM tropolone solution.

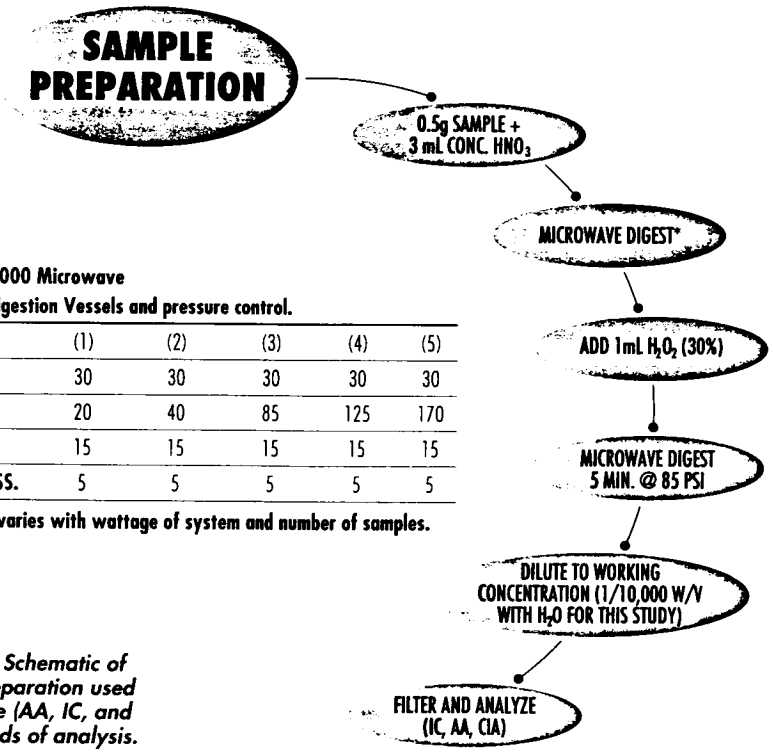
Capillary ion analysis, a relatively new technique, shows great promise for the analysis of cations from foods. Waters Capillary Ion Analysis (CIA) system is shown.

Results and Discussion

All samples were prepared using the procedure outlined in Figure 1. The amount of nitric acid used for the digestion process was minimized to 3mL. Through experimentation, we found this to be enough acid to complete the digestion without disrupting the chromatographic equilibrium during a 100 µL injection. Since the chromatography allowed the use of only 3mL of nitric acid, the samples required a secondary microwave step using H_2O_2. Moreover, sample size had to be limited to 0.5g in order for the digestion procedure to produce water-clear digests suitable for the subsequent dilutions and injections.

The Waters IC-Pak C M/D column can separate more than just the monovalent and divalent cations of nutritional importance. Figure 2 shows a separation of eight cations. This chemistry provides correlation coefficients of linearity better than 0.9996 for the range of 0-10 ppm for monovalents and 0-20 ppm for divalent cations. These ranges are representative of the analyte concentrations typically found in the final injection solution of food digests. Table 1 lists the detection limits of this ion chromatography technique as determined with low-level standard injections and based on a 3/1 signal-to-noise ratio.

The samples chosen for this study were intended to represent a wide range of the cations of interest and also a diversity of



*CEM MDS 2000 Microwave with Lined Digestion Vessels and pressure control.

STAGE	(1)	(2)	(3)	(4)	(5)
%POWER**	30	30	30	30	30
PSI	20	40	85	125	170
TIME (MIN)	15	15	15	15	15
TIME @ PRESS.	5	5	5	5	5

**%POWER varies with wattage of system and number of samples.

***Figure 1.** Schematic of sample preparation used for all three (AA, IC, and CIA) methods of analysis.*

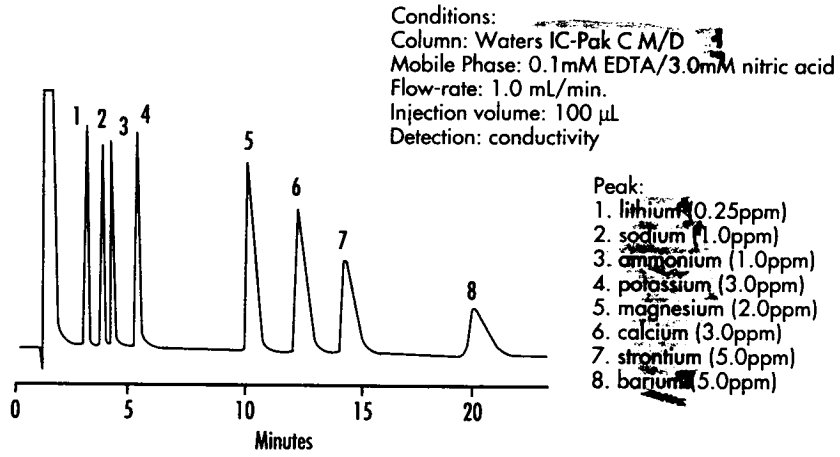


***Figure 2.** Standard separation of eight monovalent and divalent cations with coordination chromatography provides coefficients of linearity better than 0.9996 for the range of 0-10 ppm for monovalent and 0-20 ppm for divalent cations.*

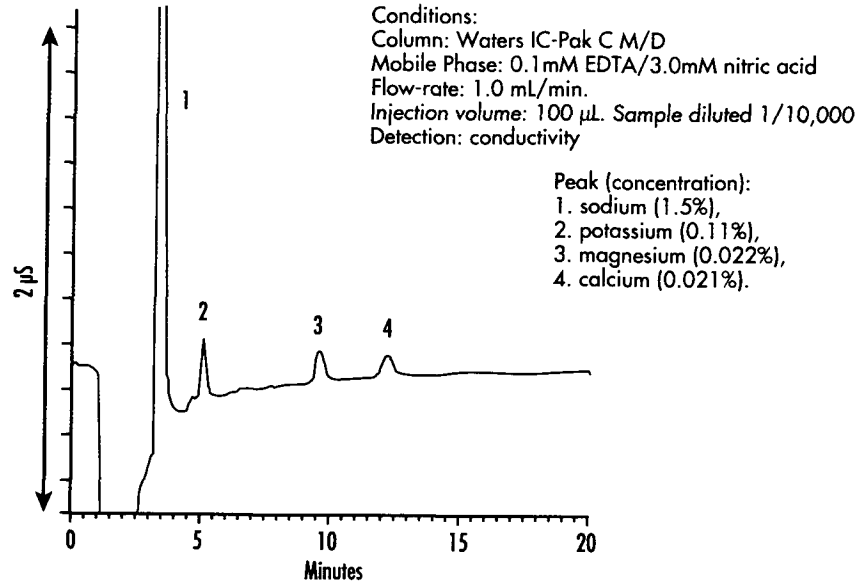


Figure 3. *Cation analysis of high sodium content (salt added) sample (pretzels) with coordination chromatography following microwave digestion.*

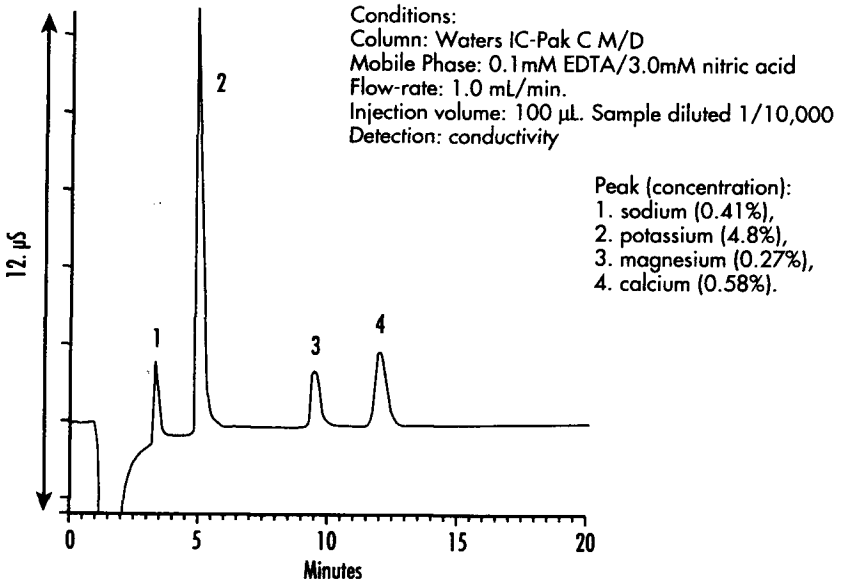


Figure 4. *Cation analysis from parsley showing much less sodium and higher levels of magnesium than other samples with coordination chromatography following microwave digestion.*

Table I.
Detection limits for monovalent and divalent cations using coordination chromatography with conductimetric detection as per Figure 2. Calculated based on a 3/1 signal-to-noise ratio of standards.

Analyte	*Concentration (ppb)*
Lithium	1
Sodium	5
Ammonium	5
Potassium	20
Magnesium	5
Calcium	10
Strontium	50
Barium	100

sample matrices. Pretzels (salted), parsley (dried), bread crumbs, Parmesan cheese and peanut butter all present different opportunities for matrix related excipients to potentially interfere with the chromatography. However, in all cases, an interference-free chromatogram was generated.

Pretzels, representing a high-sodium (salt added) content sample, generated the chromatogram seen in Figure 3. Figure 4 depicts a natural level food example using parsley as the sample. This matrix contained much less sodium. Also of interest in this example is the high level of magnesium present (0.27%) relative to the other samples. It should be noted that this is a sample high in chlorophyll, a magnesium containing compound.

Figures 5 and 6 are electropherograms generated by subjecting the IC digests of parsley and bread crumbs respectively to a CIA system optimized for cations. Compared to IC, the CIA separation is more interference-free and twice as fast.

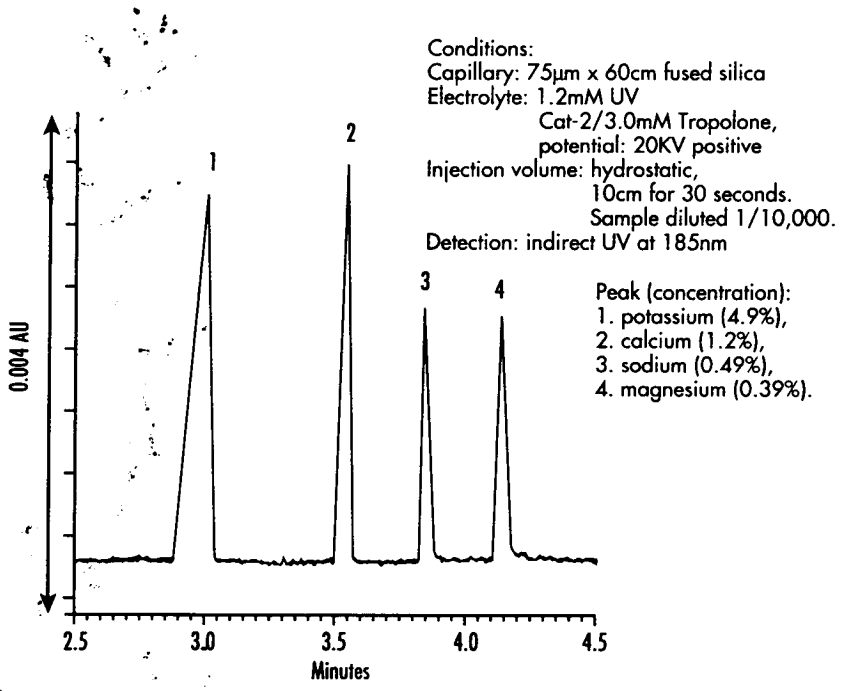


***Figure 5.** Cation analysis from parsley with capillary ion analysis following microwave digestion. The CIA separation is more interference free and twice as fast as IC.*

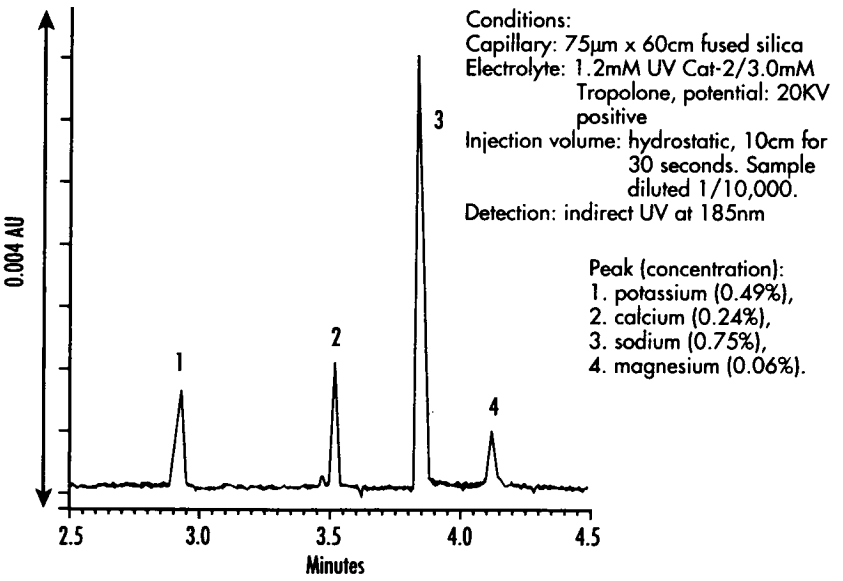


***Figure 6.** Cation analysis from bread crumbs with capillary ion analysis following microwave digestion. The CIA separation is more interference free and twice as fast as IC.*

Conclusions

Microwave digestion and either coordination ion chromatography or CIA from Waters can provide advantages over current methods of analysis for cation determinations from food. Multi-sample, multi-analyte capabilities offered by these approaches can decrease analysis time and increase throughput while minimizing sample handling. Initial ion chromatography feasibility studies with the Waters IC-Pak C M/D column have generated results which are acceptable relative to atomic absorption. CIA can also be applied to the sample digests with good results. This technique offers the advantage of speed with run times of 4.5 minutes versus 20 minutes for ion chromatography. This speed provides a better complement to fast microwave digestion sample preparation. In this example, an analytical lab incorporating microwave digestion followed by CIA can process 50 samples times 4 analytes or perform 200 cation analyses per day.

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