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On-line matrix removal of lead for the determination of trace elements in forensic bullet samples by flow injection inductively coupled plasma-mass spectrometry

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Abstract

The determination of trace elements in lead by inductively coupled plasma (ICP) source mass spectrometry (MS) is not possible without the removal of a substantial proportion of the lead matrix. This was achieved by the retention of lead from a 130-\mu l sample solution (100 mg l\textsuperscript{-1} lead in 2% v/v nitric acid) injected into a single-line (3% v/v nitric acid) flow injection manifold, on 100 mg of Pb-Spec\textsuperscript{a} packed into a cylindrical column (6 cm × 4 mm internal diameter). The analytes, Ag, As, Bi, Cd, Cu, Sb and Sn, passed through the column and were quantified against matrix-matched standards. Only Ag showed significant retention, but could still be measured in an 8-min run. The column was rinsed by flushing with 0.1 M ammonium citrate solution. Lead was monitored by flame atomic absorption spectrometry in preliminary experiments concerning column capacity and breakthrough. Although the capacity of the material in the dynamic, flow-through mode was less than the literature value based on equilibrium studies, the lead from up to 13 successive injections was sufficiently retained to allow accurate determination of the analytes without intermediate rinsing of the column. The precision [percentage relative standard deviation (%R.S.D.), \(n = 5\)] of the procedure ranged from 1.7% (100 ng ml\textsuperscript{-1} copper) to 2.8% (5 ng ml\textsuperscript{-1} cadmium), and detection limits were in the range 0.2–10 ng ml\textsuperscript{-1}. The accuracy of the procedure was assessed by the analysis of three National Institute of Standards and Technology standard reference materials (SRM 2416 bullet lead, SRM 2415 battery lead, and SRM 2417 lead base alloy). For each SRM, duplicate determinations of seven analytes were made. Of the 42 determinations, 36 fell within the confidence interval around the accepted value. Three real bullets were analyzed for seven elements by both the flow injection solid-phase extraction ICP-MS method and by aspiration of the bullet solutions (10 000 mg l\textsuperscript{-1} lead) directly into an ICP emission spectrometer. A linear least squares regression of these

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two sets of results gave a line with slope $1.01 \pm 0.04$ and an intercept of $-5 \pm 100 \mu g g^{-1}$, where the $\pm$ terms are 95% confidence intervals. The column lifetime was in excess of 5 months of daily use.

**Keywords:** Flow injection; Solid phase extraction; Lead; Matrix removal; Inductively coupled plasma-mass spectrometry

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1. Introduction

The trace element composition of bullet lead can provide important information to law enforcement personnel attempting to solve firearms-related crimes. This examination becomes even more crucial in situations where a firearm is not recovered or the crime scene bullet is too mutilated for conventional ballistics identification [1]. In these cases, the composition of the evidence bullet is compared with unfired bullets that can be associated with a suspect. The conclusion from this type of comparison is that the composition of the crime scene bullet is either analytically distinguishable or indistinguishable with that of bullets from a known source [2].

The compositional analysis of bullet lead has been performed by a variety of instrumental methods. These include neutron activation analysis (NAA), atomic absorption spectrometry (AAS), spark source mass spectrometry (SSMS), inductively coupled plasma atomic emission spectrometry (ICP-AES), and inductively coupled plasma mass spectrometry (ICP-MS) [1,2]. Each technique has its advantages and disadvantages. Neutron activation analysis and AAS are both limited by the number of elements of interest that can be determined conveniently, while the spark source and plasma methods are susceptible to interferences from the sample matrix. However, NAA and ICP-AES methods have been used extensively, particularly by the FBI Laboratory, and have been applied to thousands of cases over the past 30 years.

The multielement capability, sensitivity, and dynamic range of ICP-MS make this the ideal technique for trace element analyses of bullet lead. The most significant analytical disadvantage to using ICP-MS, however, is the interference caused by the lead in the sample matrix. Because bullets typically contain 95–99% lead [3], sample solutions must be diluted extensively to avoid the signal suppression and salt deposition resulting from the presence of the matrix. However, this dilution makes the target elements more difficult to detect. Thus, a suitable procedure would be to remove the interfering lead matrix without decreasing the concentrations of the analytes.

The selective removal of a matrix element is not a commonly performed procedure. One laboratory removed the lead from bullet digests by precipitating the Pb as its sulfate [2]. Although this off-line technique appears successful, the throughput is reduced by the requisite 1-h delay to ensure complete precipitation. Another report [4] describes the removal of a uranium matrix from a sample by utilizing the selectivity of solid-phase extraction resins. The sample was injected into a flow injection manifold containing an ion-exchange column that retained the matrix while allowing the analytes to pass through unretained.

The affinity of macrocyclic crown ethers for specific cations is well known [5–8]. Those cations with ionic radii most closely matching that of the macrocycle’s cavity will be selectively complexed by the ether. Cations that are significantly smaller than the cavity will be too far from the ring oxygens’ lone pairs to form an appreciable bond, while larger cations will not be able to fit inside the cavity at all. The selectivity of crown ethers can be used to separate certain cations from a mixture, both for preconcentration purposes and for matrix removal [9–14].

A solid-phase extraction (SPE) material containing immobilized di-t-butyl-dicyclohexano-18-crown-6 was incorporated into a flow injection manifold. This crown ether, known as Pb-Spec® is selective for and has a high capacity for lead [9,10,13]. An on-line solid-phase extraction
method has been developed for the selective re-
tention of lead from bullet samples with subse-
quent determination of Ag, As, Bi, Cd, Cu, Sb
and Sn by ICP-MS.

2. Experimental

2.1. Reagents and standards

Pb-Spec resin was obtained from Eichrom, Inc.
(Darien, IL, USA) and used as received. A stock
solution of 1000 μg ml⁻¹ Pb was prepared by
dissolving the appropriate amount of lead nitrate
in 2% HNO₃. A stock solution containing 100 μg
ml⁻¹ Sb, 10 μg ml⁻¹ Bi, Cu and Sn, 7.5 μg ml⁻¹
As, 1 μg ml⁻¹ Ag, and 0.5 μg ml⁻¹ Cd was
prepared by the appropriate dilution of 1000 μg
ml⁻¹ atomic absorption standards of the individ-
ual elements (PE Pure or Spex) with 20% HNO₃
( Optima grade, Fisher). Two working solutions
were prepared by a 100-fold dilution of the stock
with 2% HNO₃; one solution was also spiked with
Pb to a final concentration of 100 μg ml⁻¹. Cali-
bration standards were prepared by diluting the
lead-containing solution while keeping the Pb
concentration constant at 100 μg ml⁻¹. In some
instances, this required the addition of Pb from a
1000-μg ml⁻¹ Pb atomic absorption standard (PE
Pure). The carrier solution was 3% HNO₃, while
the eluent was 0.1 M ammonium citrate solution
of pH 5.23. All solutions were prepared using
deionized water with a resistivity of 18.2 MΩ
(Milli-Q, Millipore), and stored in acid-washed
polyethylene (Nalgene) bottles.

2.2. Apparatus

Two flame atomic absorption spectrometers
were used for method development. These were
both Perkin-Elmer (Norwalk, CT, USA) instru-
mments: a model 1100B with deuterium back-
ground correction, and a model 3100. Both instru-
ments were equipped with a Perkin-Elmer lead
hollow cathode lamp and were operated under the
manufacturer’s recommended conditions. The
10-cm air–acetylene burner head position was
optimized in three dimensions while aspirating a
solution containing 20 μg ml⁻¹ Pb in 2% HNO₃.
When the flow injection manifold was used for
sample introduction, the capillary of the nebulizer
was positioned in the venturi throat in such a way
that the nebulizer suction was zero. This ensured
that the pumps controlled the flow rate of the
solution entering the spray chamber. The 1100B
instrument was used in the hold mode, integrat-
ning over 5 s with 10 replicates. This produced
absorbance values spaced evenly over 50 s. The
3100 instrument was operated in the continuous
mode with an Epson LQ-850 printer for data
collection.

The sample analysis was performed with an
inductively coupled plasma mass spectrometer,
HP4500 ICP-MS (Agilent Technologies, Inc.,
Wilmington, DE, USA) equipped with a cross-flow
nebulizer and Peltier-cooled Scott-type spray
chamber. The instrument was tuned at least daily
while aspirating a solution of 10 ng ml⁻¹ Ce, Li,
Tl and Y in 5% HNO₃. The instrumental condi-
tions are listed in Table 1. The instrument’s peri-
staltic pump was used only for spray chamber
draining, and was operated at a flow rate of
approximately 2.0 ml min⁻¹. The time-resolved
mode was used, with 0.3 s residence time per
mass, and a total analysis time of 8 min. Multiple
isotopes were determined for each element.

Data from the mass spectrometer was collected
using HP ICP-MS software v.4 and processed
with HP ICP-MS Chromatographic software v.4
on a Hewlett-Packard Kayak personal computer.
Additional data analysis was performed using
PeakFit v.4.05 (AISN Software, Inc.) for the cal-
culation of peak areas. The baseline was sub-
tracted from the peaks, but the data were used
without any smoothing. This processing was done
on a personal computer using Microsoft Excel 97,
which was also used to obtain the calibration
equations by unweighted least-squares regression.

Comparison sample analysis was performed
with an Optima 3300DV inductively coupled
plasma atomic emission spectrometer (Perkin-
Elmer, Norwalk, CT, USA). Daily initiation and
calibration checks were performed according to
the manufacturer’s recommendations; samples
were arranged in the instrument’s autosampler,
sequentially aspirated into the nebulizer at a flow
Table 1  
Operating conditions for the HP4500 ICP-MS and Optima 3300DV ICP-AES instruments

<table>
<thead>
<tr>
<th></th>
<th>ICP-MS</th>
<th>ICP-AES</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rf power, W</td>
<td>1200</td>
<td>1400</td>
</tr>
<tr>
<td>Argon gas flow:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plasma gas flow rate, l min⁻¹</td>
<td>15.2</td>
<td>15</td>
</tr>
<tr>
<td>Auxiliary gas flow rate, l min⁻¹</td>
<td>0.3</td>
<td></td>
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<tr>
<td>Carrier gas flow rate, l min⁻¹</td>
<td>1.06</td>
<td>0.6</td>
</tr>
<tr>
<td>Sample and skimmer cones</td>
<td>Nickel</td>
<td></td>
</tr>
<tr>
<td>Spray chamber temperature, °C</td>
<td>2</td>
<td>Ambient</td>
</tr>
<tr>
<td>Data acquisition parameters</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mode</td>
<td>Time resolved</td>
<td></td>
</tr>
<tr>
<td>Dwell time, s</td>
<td>0.3</td>
<td></td>
</tr>
<tr>
<td>Sampling period, s</td>
<td>4.3</td>
<td></td>
</tr>
<tr>
<td>Number of points per mass</td>
<td>114</td>
<td></td>
</tr>
<tr>
<td>Total analysis time per replicate, s</td>
<td>480</td>
<td>10</td>
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<tr>
<td>Number of replicates</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Viewing height, mm</td>
<td>6.2</td>
<td>15</td>
</tr>
<tr>
<td>Isotopes or analytical wavelengths, nm</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cu⁻⁶³, Cu⁻⁶⁵, Ag⁻¹⁰⁹, Ag⁻¹⁰⁹, Cd⁻¹¹³, Sn⁻¹¹⁸, Sn⁻¹²⁰, Sb⁻¹²³, Bi⁻²⁰⁹</td>
<td>Ag/328.1, As/189.0, Bi/223.1, Cd/214.4, Cu/324.8, Sb/206.8, Sn/190.0</td>
<td></td>
</tr>
</tbody>
</table>

The flow injection manifold consisted of two peristaltic pumps (Ismatec), two six-port rotary valves, and lengths of tubing. The manifold is shown schematically in Fig. 1. The sample injection volume was 130 μl, and the SPE column was in the loop of the second valve. The three-stop Tygon pump tubing used for both pumps had an internal diameter of 1.52 mm (blue–yellow). All other tubing was 0.8 mm i.d. PTFE. The glass SPE column was 6 cm long with an i.d. of 4 mm. A complete list of the values of the relevant parameters used in the analyses is given in Table 2. The tubing lengths between the SPE column and the ICP-MS were minimized.

2.3. Method development

The purpose of this study was to investigate the feasibility of matrix removal for the compositional analysis of bullet lead by ICP-MS, so a limited optimization was carried out. Starting values for the relevant parameters were available from previous work on the behavior of Pb-Spec [9]. The ammonium citrate concentration was used as recommended [9] and remained unchanged throughout the experiments. The figures of merit selected for investigation were the lead capacity of the resin, the ability of the resin to strip lead as a function of carrier acidity, and sample volume.

Table 2  
Optimized operating conditions for the flow injection manifold

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pump tubing</td>
<td>1.52 mm internal diameter (blue–yellow)</td>
</tr>
<tr>
<td>SPE column</td>
<td>6 cm long × 4 mm internal diameter</td>
</tr>
<tr>
<td>Mass of Pb-Spec</td>
<td>100 mg</td>
</tr>
<tr>
<td>Carrier flow rate</td>
<td>1.0 ml min⁻¹</td>
</tr>
<tr>
<td>Sample volume</td>
<td>130 μl</td>
</tr>
<tr>
<td>Carrier stream</td>
<td>3% v/v nitric acid</td>
</tr>
<tr>
<td>Eluent solution</td>
<td>0.1 M ammonium acetate at pH 5.2</td>
</tr>
<tr>
<td>Eluent flow rate</td>
<td>2.6 ml min⁻¹</td>
</tr>
<tr>
<td>Elution time</td>
<td>3.0 min</td>
</tr>
</tbody>
</table>
Fig. 1. The flow injection manifold. During the inject step, pump 1 is on and pump 2 is off. The sample is injected into the carrier stream and flows through the column from left to right. During the rinse step, both pumps are on and the column is flushed with eluent from right to left.

The flow rate was set for optimum ICP-MS instrument performance and was not varied.

The glass column was packed with 100 mg of Pb-Spec resin. Glass wool in each end of the column held the material in place. The resin was initially cleaned by pumping 3% HNO₃ through it for 10 min, followed by 20 min of flushing with 0.1 M ammonium citrate. A final rinse of 3% HNO₃ for 10 min completed the process.

Initial experiments were performed to investigate the capacity of the Pb-Spec. A breakthrough curve was obtained by pumping a solution containing 1000 μg ml⁻¹ Pb in 2% HNO₃ through the column and into the 3100 FAA spectrometer. A flow rate of 1.3 ml min⁻¹ was selected. Data collection began when the solution reached the nebulizer.

The ability of the resin to retain lead as a function of the acidity of the carrier was examined. Nitric acid concentrations of 0.000%, 3.000%, 6.375% (1M), and 10.00% by volume were used. The acid content of the sample itself was also varied over a similar range [2.000%, 3.000%, 6.375% (1M), and 10.00% by volume], and the effects on the Pb peak heights were monitored by the 3100 FAA spectrometer.

The effect of the sample volume was also determined. The heights of the initial breakthrough peaks as well as the eluted peaks were measured as a function of loop size. For this measurement, 1000 μg ml⁻¹ Pb was injected into the manifold at a flow rate of 5.2 ml min⁻¹. This flow rate was selected for maximum nebulizer efficiency for the 3100 instrument.

The possibility of injecting multiple samples between column rinses was investigated. The 100-μg ml⁻¹ Pb standard containing the target elements was injected into the manifold repeatedly without subsequent elution. The elements were monitored by ICP-MS.

A qualitative experiment was performed to investigate the possibility of determining additional elements in the bullet samples. It was speculated that by removing the lead matrix without dilution, some trace elements might be detected that were previously below the detection limit of the ICP-AES method. Additional elements besides those that were determined in the quantitative procedure, but are known to be present in recycled lead, were added to the qualitative run. The elements that were included were Ag, Al, As, Bi, Cd, Cr, Cu, Fe, Ga, Ge, Hg, In, La, Ni, Sb, Se, Sn, Te, Tl and Zn. None of these elements has an ionic radius similar to that of lead [15]. The procedure was changed slightly to accommodate the increased number of elements. The dwell time was decreased from 0.3 to 0.2 s per mass. This allowed the acquisition of a similar number of points per mass as in the other runs, without generating an unmanageably large data file. No spectral interference corrections were made.

2.4. Method validation and procedures

The method was validated by the analysis of NIST reference materials (SRM 2416, bullet lead; SRM 2415, battery lead; and SRM 2417, lead base alloy) and three bullet samples that were prepared by the FBI Laboratory using its current procedure used for ICP-AES. Briefly, an approximately 60-mg sample from the interior of the bullet was weighed to the nearest 0.01 mg. The sample was flattened between sheets of plastic and placed in a 15-ml polypropylene centrifuge tube. Six milliliters of 20% HNO₃ were added to the sample, and the capped tube was irradiated in
a microwave oven for 40 s on full power. If the sample mass deviated from 60 mg, the volume of the 20% HNO$_3$ was adjusted accordingly to maintain a 10.00-mg ml$^{-1}$ dilution factor. The sample was then transferred to a conventional oven and heated at 80°C for a minimum of 4 h or until digestion was complete. Ten microliters of 49% HF were added to the cooled tube followed by vortex mixing. The sample was allowed to sit overnight to ensure complete dissolution. The digest was analyzed by ICP-AES using direct aspiration. The unused portion of the same digest then underwent a final 100-fold dilution with 2% HNO$_3$ immediately prior to analysis by ICP-MS.

Sample and standard solutions were injected into the flow injection sample introduction system shown in Fig. 1. Pump 1 was always on and operating with a flow rate of 1.0 ml min$^{-1}$. In the sample inject mode, pump 2 was off. The data collection was synchronized to begin with the switching of the injection valve. The carrier solution of 3% HNO$_3$ flushed the sample through the Pb-Spec column and into the spectrometer. Once the data collection was completed, the second pump was turned on and the SPE valve turned to the rinse position. The eluent, 0.1 M ammonium citrate, backflushed the column for 3 min at a flow rate of 2.6 ml min$^{-1}$. During the elution step, the carrier continued to flow into the ICP-MS. When the elution was complete, the valve was returned to the load position and the column was rinsed with the carrier in preparation for the next injection.

Calibration was performed by injecting multi-element standards containing matrix-matched lead and HNO$_3$ concentrations. Samples and standards were analyzed in triplicate.

3. Results and discussion

3.1. Variation of system parameters

The mass of Pb-Spec packed into the column was set at 100 mg by preliminary calculations. According to Horwitz et al. [9], the resin capacity is 20.5 mg of Pb per ml of bed volume. For a bed density of 0.37 g ml$^{-1}$, the capacity is 55.4 mg Pb retained per gram of resin. The sample digestion procedure initially produces a solution containing 10 000 µg ml$^{-1}$ Pb. Using this concentration and the published capacity of the column, 100 mg of resin should strip the lead from 0.55 ml of sample. This is a common sample volume used in flow injection analysis, so the mass of resin was considered appropriate. The samples were diluted from 10 000 to 100 µg ml$^{-1}$ Pb so that breakthrough would not accidentally take place.

The Horwitz capacity [9] was based on an off-line measurement, but the working capacity of the resin could be significantly different for an on-line experiment. The dynamic capacity of the Pb-Spec resin was evaluated by acquiring a breakthrough curve, as shown in Fig. 2. The absorbance signal began to increase from its baseline value at 65 s and did not reach its maximum value until well after 2 min. This instrument was operated at a flow rate well below its optimum value for maximum sensitivity, and the detection limits of FAAS are much higher than those of ICP-MS. In principle, the column effluent could have been monitored by ICP-MS, but the potential damage to the instrument when the lead broke through the resin prohibited the use of ICP-MS for this experiment. The characterization of the breakthrough curve prior to approximately 65 s was, therefore, not possible. The sharp rise in lead concentration associated with breakthrough would have been detected at the same time, i.e. after 2 min of constant sample introduction regardless of the instrument that was used. Based on this figure, the resin stripped approximately 1.5 mg Pb from 1.5 ml of solution, corresponding to 15 mg Pb per gram of resin. A comparison of this value to the Horwitz value of 55.4 mg Pb for the same mass of resin (see above) indicates the effect of conversion to a flow-through system. Although the on-line capacity is lower than the comparable off-line value, the retention of 1.5 mg of lead is quite satisfactory for the application. Any potential signal suppression caused by the lead that bled through the column before actual breakthrough was examined by ICP-MS in the repeated injection experiment and will be discussed later.

To examine the acid dependency of the metal sorption on the resin, the acid content of both the
The curve is the average of three replicate measurements.

sample and the carrier solution were varied. According to Horwitz et al. [9], the lead sorption peaked at an acid concentration of approximately 1 M HNO₃. The acid concentrations of the sample and carrier solutions were thus distributed around 1 M. Fig. 3 shows the results. For each sample, the measured absorption was greatest with a carrier acidity of 3%. Though it is possible that the maximum value is achievable at a lower acidity than the minimum tested in this experiment, 3% was used for the remainder of the study. The differences in absorption for the sam-

Fig. 2. Breakthrough curve for 1000 μg ml⁻¹ Pb at a flow rate of 1.3 ml min⁻¹. The curve is the average of three replicate measurements.

Fig. 3. The effect of varying acid content of both the carrier solution and sample. Samples were individually prepared by dissolving the appropriate amount of lead nitrate in nitric acid. Lead concentrations for each sample were: 2% HNO₃, 1000 μg ml⁻¹; 3% HNO₃, 974 μg ml⁻¹; 6.375% HNO₃, 971 μg ml⁻¹; 10% HNO₃, 972 μg ml⁻¹. The legend lists the sample acidities as percentages.
ple acidity, however, do not match the trend observed in the case of the carrier. Although the highest absorption was achieved for sample acidities of 3% and 10% HNO₃, all values gave acceptable results, so a value of 2% was selected for the remainder of the experiments. The gain in signal intensity for the 3% and 10% samples was not deemed to be sufficiently great to warrant using these concentrations, since maintaining a simple sample preparation procedure was more important.

The effect of sample volume on the results is shown in Fig. 4. The volume of the 1000-µg ml⁻¹ sample injected had a significant effect on the efficiency of the lead retention. The peak height absorption of the breakthrough peaks increased with sample volume, as did the absorption of the elution peaks. Although more lead was retained when larger volumes were injected, the amount of lead bleeding through the column increased as well. The only volume for which the breakthrough absorption is zero is the smallest volume, 130 µl. This suggests that for volumes larger than 130 µl the residence time in the column is too short to ensure total retention. Although 130 µl was selected as the sample volume for the remainder of the study, it is possible that larger volumes could in fact be used. The amount of lead that breaks through the column may not be enough to cause signal suppression for the target analytes. Again, the choice of value for this parameter involves a compromise: the amount of lead breaking through the column compared to the potential signal augmentation from using a larger sample volume. For this study, minimum lead breakthrough was more important because the detection limit of the ICP-MS was adequate for the determination of the target analytes, even after the dilution caused by dispersion processes in the flow injection manifold.

Further experiments are necessary, however, to characterize the relationship between sample volume and the magnitude of the breakthrough. It is unclear whether this phenomenon is related to the kinetics of the flow-through system, in which case the residence time of the sample within the column is important to ensure complete lead retention. This would suggest that a slower flow rate or a longer, thinner column design would result in more complete adsorption. Another possible explanation may involve the surface characteristics of the resin. In this case, the initial portion of the sample may react with the most easily accessible crown ether cavities, shielding the remaining cavities from the rest of the sample. The column capacity that was observed in the breakthrough experiment suggests that for 1000-µg ml⁻¹ solutions, sample volumes much larger

![Graph](image1.png)

**Fig. 4.** The effect of changing sample volumes on the heights of breakthrough as well as elution peaks. The top line shows the elution peak heights, and the lower line shows the heights of the breakthrough peaks. The error bars represent ±1 S.D. of the multiple replicates. PE 3100 FAAS detection, 5.2 ml min⁻¹.
than 130 µl could be tolerated by the resin. If the column can really retain 1.5 mg of lead from a flowing stream, then the injection of up to 1500 µl should not result in breakthrough. This inconsistency could be caused by the different flow rates used for the breakthrough curve (1.3 ml min\(^{-1}\)) and the sample volume experiments (5.2 ml min\(^{-1}\)). Research is currently underway to answer some of these questions.

3.2. Analytical performance

The flow injection solid-phase extraction system was used to remove the lead matrix from a series of calibration standards. Linear calibration plots were fitted to the points generated by the three standards plus a blank using an unweighted least-squares algorithm. The results are given in Table 3. Because the results did not differ significantly between isotopes of a single element, only one isotope is listed for each element. The limit of detection for each element was calculated [16] as the concentration giving a signal equal to three times the root mean square error of the y-residuals in the regression analysis.

The limits of detection are in the low ng ml\(^{-1}\) range. While these values are perfectly adequate for the analysis of bullets, they are higher than typically observed for direct nebulization sample introduction using this ICP-MS instrument. The increased limits of detection may be a consequence of the dilution that occurs in the manifold. One estimate for the on-line dilution, generated by the comparison of the count rate for direct nebulization of an element with its peak maximum value, is approximately 30-fold. A second estimate, derived from a comparison of the injected volume to an estimated peak volume of 2.0 ml based on the peak basewidth and flow rate, gives a value of approximately 15-fold. A dilution factor of 15–30 may explain the limits of detection that, although adequate for this application, were rather high. These values may be improved by decreasing the amount of sample dilution in the system.

Because the Pb-Spec resin has such a high capacity for lead, injecting samples repeatedly without an intermediate elution step is possible. This would increase sample throughput, since the time required for elution and column rinsing between replicate measurements could be eliminated. The potential disadvantages could be memory effects or signal suppression caused by lead bleeding through the column before breakthrough. The \(^{208}\)Pb signals for repeated injections are shown in Fig. 5. The lead background increased with each injection, demonstrating that although the column was retaining the lead some column bleed occurred. Fig. 6 shows the effect of repeated injections on the \(^{107}\)Ag peak, which was used here as a model for the other elements. As the number of injections increased, the \(^{107}\)Ag peak remained essentially unchanged. Minor variations in the peak appearance occurred, but the R.S.D. (\(n = 5\)) of the peak area was less than 3.2\%. Similar peak area precisions were observed for the other elements. This demonstrates that the amount of lead bleeding off the column was not

<table>
<thead>
<tr>
<th>Element</th>
<th>Slope(^a) (counts ng(^{-1}) ml)</th>
<th>Intercept(^a) (counts)</th>
<th>Correlation coefficient</th>
<th>LOD (ng ml(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>(^{63})Cu</td>
<td>446 ± 33</td>
<td>330 ± 2100</td>
<td>0.9997</td>
<td>4</td>
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<tr>
<td>(^{75})As</td>
<td>91 ± 7</td>
<td>43 ± 330</td>
<td>0.9997</td>
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<tr>
<td>(^{107})Ag</td>
<td>426 ± 61</td>
<td>−68 ± 390</td>
<td>0.9989</td>
<td>0.8</td>
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<tr>
<td>(^{111})Cd</td>
<td>97 ± 7</td>
<td>14 ± 23</td>
<td>0.9997</td>
<td>0.2</td>
</tr>
<tr>
<td>(^{119})Sn</td>
<td>242 ± 2</td>
<td>130 ± 150</td>
<td>1.0000</td>
<td>0.6</td>
</tr>
<tr>
<td>(^{121})Sb</td>
<td>240 ± 6</td>
<td>−713 ± 3800</td>
<td>0.9999</td>
<td>10</td>
</tr>
<tr>
<td>(^{209})Bi</td>
<td>429 ± 55</td>
<td>540 ± 3500</td>
<td>0.9991</td>
<td>7</td>
</tr>
</tbody>
</table>

\(^a\)The ± terms are 95% confidence intervals calculated according to the procedure described by Miller and Miller [16] for four calibration points, each of which is the mean of three replicate measurements.
sufficient to cause signal suppression of the target analytes. Although the number of injections was halted at 13, additional injections could have been made. The breakthrough point of the column was not attained, since the Pb signal did not increase as shown in Fig. 2. Each injection of 130 μl of 100 μg ml⁻¹ Pb contained 13 μg of Pb. The column was capable of retaining 13 injections, or 169 μg of Pb. Because the column did not reach breakthrough, the working capacity of the column is greater than 169 μg. The working capacity of the column may be as high as its calculated capacity of 1.5 mg, but it is possible that the lead bleeding from the column would be enough to cause signal suppression before the column capacity is reached.
The peaks obtained by the flow injection SPE technique for most elements had basewidths of approximately 1.5 min. The Ag peaks were broader, having basewidths of nearly 6.5 min. This is illustrated in Fig. 7, which shows the overlay of one $^{107}$Ag peak from Fig. 6 with the corresponding $^{63}$Cu peak from the same sample injection. The Ag peak basewidth is approximately four times greater than that of the Cu peak. Although the peak areas were reproducible, the time required for the analytes to pass through the system was long, resembling a typical chromatography experiment rather than a flow injection one. The peaks for most analytes in the sample analyzed were easily integrated, but on occasion, some elements were below the limits of detection. By decreasing the broadening, it may be possible to determine these elements. Further developments of the system are in progress, including an examination of the effect of column size.

One column was used daily for 5 months, during which time several hundred injections were made, without any loss of performance. Column lifetime was not determined in this study because it was never reached.

3.3. Results for samples

The results of the analysis of three SRMs are shown in Table 4. Two samples of each SRM were separately digested and analyzed in triplicate. The elemental concentrations are expressed as the mean together with the 95% confidence interval for each value in parentheses. The concentrations that were calculated for each isotope of a specific element did not differ significantly from one another, so results for only one isotope are shown for each element. The value of the uncertainty for the SRMs, also shown in parentheses, is the value from the certificate. Since this value is estimated, it is impossible to perform any statistical test to determine whether the concentrations are significantly different. As an approximate basis for judgment, however, the concentrations can be assumed to be indistinguishable if the mean of one falls within the confidence interval about the other. Twenty-eight of the possible

![Fig. 7. The peaks for $^{63}$Cu and $^{107}$Ag from the first of the repeated injections. The signal for $^{63}$Cu has been divided by a factor of 20 to match the scale of $^{107}$Ag. The width of the silver peak is approximately four times that of the copper peak.](image-url)
Table 4
Concentrations of the trace elements in NIST standard reference materials after lead removal

<table>
<thead>
<tr>
<th>Element</th>
<th>SRM 2416, Bullet Lead</th>
<th></th>
<th>SRM 2415, Battery Lead</th>
<th></th>
<th>SRM 2417, Lead-based Alloy</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Certified</td>
<td>Sample A</td>
<td>Sample B</td>
<td>Certified</td>
<td>Sample A</td>
</tr>
<tr>
<td>Cu</td>
<td>650 (20)</td>
<td>660 (60)</td>
<td>640 (20)</td>
<td>950 (50)</td>
<td>940 (30)</td>
</tr>
<tr>
<td>As</td>
<td>560 (10)</td>
<td>540 (20)</td>
<td>520 (30)</td>
<td>2000 (100)</td>
<td>1900 (40)</td>
</tr>
<tr>
<td>Ag</td>
<td>44 (2)</td>
<td>39 (3)</td>
<td>35 (3)</td>
<td>20 (10)</td>
<td>23 (4)</td>
</tr>
<tr>
<td>Cd</td>
<td>2</td>
<td>1.1</td>
<td>1</td>
<td>20 (10)</td>
<td>21.3 (0.4)</td>
</tr>
<tr>
<td>Sn</td>
<td>900 (100)</td>
<td>730 (110)</td>
<td>700 (40)</td>
<td>3300 (100)</td>
<td>2830 (250)</td>
</tr>
<tr>
<td>Sb</td>
<td>7900 (100)</td>
<td>7620 (370)</td>
<td>7500 (140)</td>
<td>29500 (400)</td>
<td>28280 (800)</td>
</tr>
<tr>
<td>Bi</td>
<td>1000 (100)</td>
<td>990 (50)</td>
<td>920 (50)</td>
<td>540 (40)</td>
<td>537.9 (0.6)</td>
</tr>
</tbody>
</table>

Concentrations are in mg g\(^-1\) in the solid sample. Values in parentheses are 95% confidence intervals for three replicate measurements for the experimental values, but are estimated uncertainties in the certified values. ND: Not detected.

\(^{a}\) Reference value, not certified.

\(^{b}\) Value obtained through repeated measurement, not certified.
42 comparisons met this criterion and for a further six comparisons, the confidence intervals overlapped. For eight comparisons, the confidence intervals did not overlap. The element with the highest degree of inaccuracy was tin.

The method was also used to determine the trace elements in some real samples. Table 5 gives the results for the analysis of three bullets by both ICP-MS and ICP-AES. A two-point calibration curve was used, with SRM 2416 as the high concentration for each element and a blank solution as the low concentration. A two-point calibration is standard procedure for the FBI Laboratory’s ICP-AES method, so a similar calibration was applied to the ICP-MS method. The ICP-MS calibration used a 3% HNO₃ solution as the blank. The ICP-AES procedure used a high-purity lead solution as the low concentration standard for the target elements. Table 5 shows that the elemental concentrations obtained by both techniques are similar. A regression analysis in which the ICP-MS data were plotted vs. the ICP-AES data gave a line of slope 1.01 ± 0.04 and intercept −5 ± 100 µg g⁻¹. The uncertainties represent the 95% confidence interval [16]. It is possible that some of the differences in individual concentrations are caused by the use of different blank solutions in the calibration. Of the 21 possible comparisons of precisions, there are no significant differences between the ICP-AES results and those of the ICP-MS method (F-test, 95% confidence) for 16 of the comparisons. The differences that were observed could be the result of instrumental drift: triplicate emission measurements were made over a 30-s period, whereas the triplicate mass spectral measurements were made over approximately 24 min.

To evaluate the possibility of additional discrimination capability provided by increasing the number of analytes, the digest of bullet Q1a was analyzed qualitatively by ICP-MS. The elements currently utilized for bullet lead characterization were selected based on ICP-AES and NAA studies. The source discrimination capability of each element is a function of a number of factors including homogeneity within a source, variability across sources, and precision of the analytical method. One potential advantage of ICP-MS as a detection method is that additional elements may fit the third of these criteria, i.e. can be determined with good analytical precision.

As ICP-MS is a rapid sequential measurement system, increasing the number of elements measured has consequences in terms of the number of points per peak collected. However, if 20 points per peak are taken to give good accuracy and precision (i.e. absence of spectral skew), then for a combined settling and dwell time of approximately 50 ms up to 60 isotopes could be measured for a peak of 60 s basewidth [17].

Of the 20 additional elements (Ag, Al, As, Bi, Cd, Cr, Cu, Fe, Ga, Ge, Hg, In, La, Ni, Sb, Se, Sn, Te, Tl and Zn) that were tested in the qualitative run (15 points per peak), only Al, Ga, La, Ni and

<table>
<thead>
<tr>
<th>Element</th>
<th>K1a</th>
<th>K2a</th>
<th>Q1a</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>SPE-ICP-MS</td>
<td>ICP-AES</td>
<td>SPE-ICP-MS</td>
</tr>
<tr>
<td>⁶³Cu</td>
<td>400 (28)</td>
<td>404 (8)</td>
<td>554 (15)</td>
</tr>
<tr>
<td>⁷⁵As</td>
<td>31 (10)</td>
<td>21 (1)</td>
<td>33 (2)</td>
</tr>
<tr>
<td>¹⁰⁰⁷Ag</td>
<td>21.9 (0.1)</td>
<td>26.2 (0.3)</td>
<td>26 (1)</td>
</tr>
<tr>
<td>¹¹¹⁷Cd</td>
<td>0.1, 0.6⁶</td>
<td>0.25 (0.01)</td>
<td>1 (2)</td>
</tr>
<tr>
<td>¹¹⁵Sn</td>
<td>16 (9)</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>¹²³⁷Sb</td>
<td>8561 (686)</td>
<td>8323 (150)</td>
<td>6632 (340)</td>
</tr>
<tr>
<td>²⁰⁹⁰Bi</td>
<td>100.8 (0.6)</td>
<td>93 (6)</td>
<td>106 (2)</td>
</tr>
</tbody>
</table>

Concentrations in mg g⁻¹ in the solid sample. Values in parentheses are 95% confidence intervals for three replicate measurements of each lead digest solution. ND: Not detected.

⁶ Two replicates.
Zn were detected in the bullet. Each of these elements was detected as a discernable peak, although the role of spectral interferences has yet to be investigated. The Ga peak was broader than the others, suggesting that Ga was slightly retained by the column, despite the fact that its radius at 62 pm is smaller than that of Pb at 119 pm [15]. It is possible that the procedure is capable of detecting additional elements in bullets that were not present in this specific sample. For example, Keto [2] reports the successful determination of Te in NIST SRM 2416. Further studies are needed to determine the resin retention characteristics of any additional elements, as well as their variation among lead sources.

4. Conclusions

It is concluded that it is possible to remove the high concentration of lead in bullet digests by on-line, solid-phase extraction with Pb-Spec to an extent that allows determination by ICP-MS of the trace elements commonly used in forensic comparisons. This suite of elements could be extended to include Al, Ga, La, Ni and Zn, although the forensic utility of these additional elements has yet to be determined. The procedure developed is not yet fully optimized, as the factors governing the dynamic capacity of the column have not been unambiguously resolved and the dispersion in the manifold could be reduced, thereby decreasing peak widths and improving the signal-to-noise ratio. Further studies of these factors are in progress. It is anticipated that the performance of an SPE-ICP-MS procedure will ultimately complement that of the current ICP-AES method.

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