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A New Method for Collection and Identification of Gunshot Residues from the Hands of Shooters*

ABSTRACT: This work presents a novel collection method for gunshot residues (GSR) using a sampling procedure based on ethylenediaminetetraacetic acid (EDTA) solution as a complexing agent on moistened swabs. Detection was via a sector-field inductively coupled plasma mass spectrometry (HRICP-MS). The proposed collection and analytical method allowed detection of antimony (Sb), barium (Ba) and lead (Pb) after .38 shot tests, at detection limits of less than 1 $\mu g\ L^{-1}$ in four different areas of the hands of volunteers. This paper includes a discussion concerning hand areas near the thumb and forefinger as being more suitable for GSR collection as well as a comparison between differences observed using 2% diluted EDTA, 2% nitric acid solution, and simple deionized water as collecting solutions, proving the superior efficiency of EDTA in GSR recoveries.

KEYWORDS: forensic science, gunshot residues, collection, barium, lead, antimony, inductively coupled plasma-mass spectrometry

In criminal investigations, analyses of GSR are usually required to determine whether or not a suspect discharged a firearm, to confirm a bullet entrance hole or to estimate a firing distance. In these studies, a police investigation demands a positive identification of GSR through chemical analysis on the suspect's clothes, hair and in most cases, his hands (1–4). In general, these techniques involve determination of organic or inorganic compounds encountered in the propellant and primer, respectively. Previous studies (1) reported in forensic laboratories for GSR samples consider antimony, lead, and barium as major elements present on GSR particles. In consequence, the detection of these elements is of great importance in criminal investigations.

Different methods have been used successfully in forensic laboratories for GSR analysis, each exhibiting many advantages and drawbacks (5,6). Neutron activation analysis (NAA) has been used since the 1960s, even though it was not applicable to lead, and few laboratories had the required high-flux neutron sources (3). The application of conventional flame atomic absorption spectroscopy (AAS) started in the early 70's, with sensitivity for barium and antimony equivalent to that obtained by NAA. AAS required relatively simple and inexpensive instrumentation (compared to that required for NAA) and had sufficient sensitivity for lead detection (1,3,7). In a 1971 report, Krishnan encountered lead concentrations in the range of 1.3 to 7.6 μg for a non-firing hand and 5.2 to 30.0 μg for a firing hand, but conventional flame AAS used was found inadequate for barium and antimony at the levels found in GSR swab extracts (2,8). The introduction of electrothermal analyzers (GFAAS), proposed by Newton in 1981 (9), eliminated such inadequacies, but the analyses were relatively time-consuming and subject to non-analyte interferences, as reported by Koons (10). Moreover, NAA and AAS (GFAAS) determine only the total amount of each element, with no indication whether they are present as metal or in a particular combined form (3), information which might allow the analyst to verify whether the residues detected were derived from a shot. As a consequence, scanning electron microscopy/energy X-ray spectroscopy (SEM/EDX) was considered from the mid-1970s, as a technique to examine the morphology and the chemical composition of micron-size particles in a non-destructive way, as reported in many studies (1–4,11–15). In SEM/EDX, however, it was necessary to locate GSR particles, increasing the analysis time so that its use was restricted to special cases. The development of automated systems improves efficiencies but does not overcome the long sample throughput time, with some samples taking up to 8 h, as reported by Tillman (12). Inductively coupled plasma emission spectroscopy (ICP/AES) has gained acceptance as a rapid technique with capabilities for multielemental analysis relatively free from interferences. Koons et al. (16), compared this technique with AAS for barium measurements in swab extracts, reporting a superior precision and accuracy for the determination of Ba, the lack of Ba background emission, low detection limits and a wide linear dynamic range as advantages for ICP-AES. However, ICP-AES commercial instruments lack the sensitivity required for accurate Sb determination on GSR swab extracts (16).

Inductively coupled plasma mass spectrometry (ICP-MS) (17,18) has emerged over the last two decades as a technique with many additional analytical benefits for GSR studies, allowing a reliable determination of analytes, particularly in trace analysis. The analytical detection limits are superior to those obtained from other techniques, with a multi-element capability with the accuracy and precision required to GSR analysis. Koons (19) reported the use of ICP-MS for analysis of GSR swab extracts, obtaining detection limits corresponding to 0.5 ng of Sb, 0.2 ng of Ba, and 1.4 ng of Pb,

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without some limitations observed in other techniques, such as spectroscopic interferences. Nevertheless, few reports about ICP-MS applied to GSR studies have been made. The relatively high cost of analysis may be responsible for its limited use, but this highly sensitive and selective method for elemental and isotopic analysis can be a powerful tool for forensic scientists to deal with diverse matrices.

One of the main steps in GSR analysis consists of the collection of particulate material from the hands of suspects to determine elements of interest. Some techniques are generally used for a complete removal of GSR particles, such as vacuum and tape lifting and swabbing (1,20). Vacuum lifting is more appropriate for sampling clothes, while the loss of stickiness in tape lifting can result in a reduced sampling efficiency. On the other hand, swabbing methods permit a significant removal of GSR particles. In swabbing, to recover as much GSR as possible, a selected area of the hand is scrubbed with cotton moistened with an appropriate solvent. Repeated swabbing increases the efficiency of extraction and can transfer most of the GSR found on the surface of the skin, even those present in wrinkles. The recovery of analytes by solvents can also be effective considering that elements found after a firegun discharge may be present not only in metallic form but also in ionized form, derived from the gaseous discharge associated with the burning of the primer (21). The presence of ionized analytes facilitates the chemical extraction of elements on the surface of the hands using different solvents (1,7), such as diluted acids for inorganic GSR sampling, even though these cause some irritation to the skin.

The collection method developed in this paper considered moistened swabs with ethylenediaminetetraacetic acid (EDTA), a powerful and well-known complexing agent. The EDTA molecule contains a sufficient number of ligand groups to permit bonding with all coordination positions of a metal ion, forming 1:1 complexes with many metals, regardless of oxidation state. The ability of EDTA to form anionic metal complexes greatly alters the oxidation-reduction potential of the metal ion and also enhances the partitioning of the metal to the aqueous solution. Depending upon the exact conditions of the application, EDTA is strongly attracted to alkaline-earth and transition metal ions at surfaces to form water soluble metal-EDTA chelates, converting all di- and tri-valent metallic ions to an anionic form, besides acting as depressor in any pH (although best at 3.5 to 7.5) for long periods of time (22,23). Barium complexes, for instance, present a complete dissociation at low pH (about 2–3), while lead presents more stable complexes in basic solutions. Related reports describe EDTA uses for Sb (24,25) in many situations and techniques, even though information about organoantimony species is scarce. However, the solubility of antimony pentoxide in water and aqueous solutions must be considered. During water bath digestion, EDTA can form salts with nitric acid (a stronger acid) by virtue of its amino groups, but strong heating of the acid alone causes decarboxylation and complete breakdown of the molecule, separating the chelating agent from the metal. A comparison of the efficiencies of EDTA, diluted nitric acid and water as collection solutions, is presented in this work, indicating the benefits of the use of EDTA in the recovery of GSR deposits from hands. Different hand areas were studied and compared by the analysis of extract solutions using HRICP-MS (26).

Materials and Methods

The test shots were undertaken at the Ballistic Laboratory of the São Paulo Criminal Institute (I.C.-SP), in order to evaluate the proposed method for GSR detection in different areas of the hand. The choice of Taurus and Rossi handguns caliber .38 as well as CBC (Cia Brasileira de Cartuchos) .38 SPL LRN (lead round nose) cartridge case was based on São Paulo Police apprehension firearm statistics, which show that caliber .38 handguns and lead cartridges represent about 50% of the total. The shot tests were executed with different .38 weapons obtained from police apprehensions, and involved male and female volunteers chosen at random and who did not generally handle firearms. In a period up to 60 min after a .38 handgun discharge, a moistened swab with 2% EDTA solution was applied to four specific areas on the surface of the shooter's hand, including palm, back, thumb and forefinger palm, and the back of the thumb and forefinger, as indicated in Figs. 1*a* to 1*d*. ^{121}Sb, ^{138}Ba and ^{208}Pb isotopes were analyzed for determination of total analytes concentrations. A hand blank test was also made on each volunteer before the shot tests. Moreover, the weapons were cleaned prior to each test shot to eliminate any possibility of contamination from previous discharges. In addition, deionized water of 18 MΩ quality and diluted nitric acid solution were applied to GSR collections undertaken from the thumb and forefinger regions of the hand, as mentioned in a previous work (27).

Initial tests compared the GSR recoveries obtained using deionized water, 2% diluted nitric acid and 2 and 5% EDTA disodium as collection solutions, where 32 test shots were made for each collection solution studied. The collection tests with nitric acid were conceived to be applied in concentrations minimally damaging to the skin. A 2% EDTA solution used in this method guaranteed an extraction performed at pH 4.5 to 5.0, which is consistent with

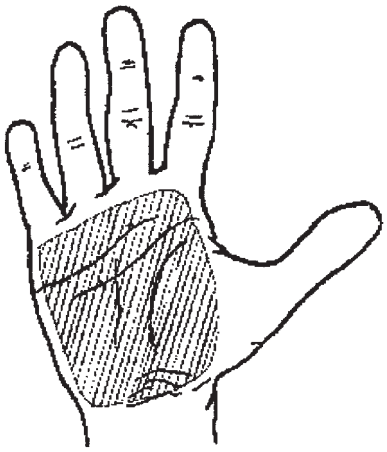

FIG. 1*a*—*Palm area.*

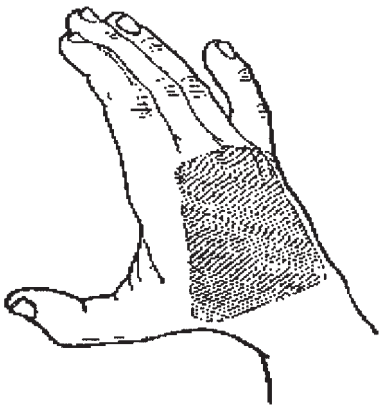

FIG. 1*b*—*Back area.*

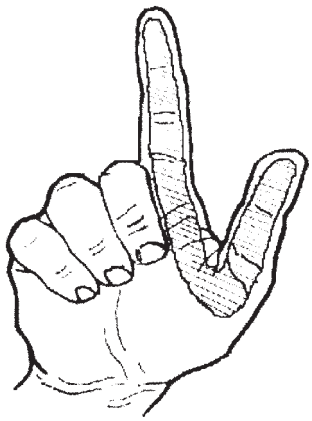

FIG. 1*c—Thumb and forefinger palm area.*

FIG. 1*d—Thumb and forefinger back area.*

EDTA's ability to form chelates with all di- and trivalent metal ions, in any dilution at pH at 3.5 to 7.5. Previous collections performed with 5% EDTA solution revealed no differences with the results of collections performed in the same way with 2% EDTA solution, justifying the use of 2% EDTA. The apparatus was composed of a flask (containing an appropriate collection solution), a package containing some swabs, covered 10 mL polypropylene tubes (SARSTEDT, Germany) and a pair of scissors. The collection method consisted in moistening swabs in the solution for about 2 min, and scrubbing around and behind the thumb and forefinger for 1 min. The cotton swab was then sectioned by using the scissors (washed between each sample) and put inside the polypropylene tube, which was covered, identified and taken to the laboratory. The samples were then submitted to digestion with 2 mL of a 10% nitric acid (65% Suprapur MERCK, Germany) solution, followed by 5 min agitation at 25 KHz in an ultrasonic bath (UNIQUE, Model TA1800, Brazil) and 1 h in an 80°C water bath (7). Afterwards, extract sample solutions were diluted to 10 mL with deionized water and aspirated directly into a sector-field inductively coupled plasma mass spectrometer (ELEMENT 1, Finnigan MAT, Bremen, Germany) for the determination of Sb, Ba and Pb. Working standard solutions containing 1, 5, 10, 50, 100, 200 and 300 $\mu g\ L^{-1}$ at 1% nitric acid were prepared by dilution of Ba, Sb and Pb original 1000 $\mu g\ L^{-1}$ SPEX standards (NJ). A Meinhard concentric nebulizer was used for sample introduction to a quartz torch, with peristaltic pumping, and 10 $\mu g\ L^{-1}$ of ^{115}In solution (SPEX) was used to verify the sensitivity during the analysis sequence. The main operation conditions are given in Table 1.

Results

An initial comparison of GSR recoveries among different collection solutions, verified the analytical advantages of using diluted 2% EDTA, as can be observed from Table 2. For this specific group of people, it is observed that the maximum values of the analytes in blank tests are greater than the minimum recoveries observed after shot tests. In addition, the results are not regularly distributed (especially for Ba and Pb recoveries), as shown by differences observed between median and mean values. In real situations, where recoveries before a shot occurrence are not possible, the regular activities of the suspect should be considered (11).

Table 3 presents a statistical summary of concentration data of samples obtained from the hands of volunteers before and after shot firing. Antimony, Ba and Pb recoveries from the hands of shooters are greater on areas near the thumb and forefinger (TF-Palm and TF-Back), suggesting that these specific areas are more suitable for GSR collection. Antimony recoveries showed this clearly in the data of Fig. 2, especially considering that Sb is uncommon in the environment and the workplace; high concentrations of antimony detected on hands, combined with high concentrations of lead detected from this hand area is a strong indication of the use of a .38 firearm. Observation of the data presented in Table 3, however, reveals a random distribution of elemental concentrations about their mean values, which makes a general interpretation of the results difficult. As also observed from Table 2, these wide variations can be seen not only after but also before test shots, so that the mean values carry no significance, especially when compared with detection limits obtained for the analytes: detection limits, defined as three times the base noise levels, of 0.045 $\mu g\ L^{-1}$ for antimony, 0.507 $\mu g\ L^{-1}$ for barium and 0.117 $\mu g\ L^{-1}$ for lead, were obtained with a 2% diluted EDTA collecting solution. However, all the results obtained from all hand areas after shot tests were found to be distributed within 45% of median values, showing a characteristic pattern. Thus, even though recoveries of analytes revealed variations in concentrations, a clear distribution is observed around the median values. Considering that all of the arms were obtained in police apprehensions, the wide dispersion of analyte recoveries from different shooters can possibly be attributed to causes such as those mentioned previously; namely environmental and occupational influences, the shape and size of the hands, and particularly the state of preservation of the firearms. In addition, the natural pH of the skin may represent another variable key; as mentioned in previous studies (29), pH range of the skin may vary from 4.0 to 7.0, due to low molecular weight fatty acids and volatile aliphatic acids originating from sweat and sebaceous glands together with lipids from degraded cell cytoplasm. These characteristics com-

TABLE 1—*HRICP-MS main operating conditions.*

Cool gas flow rate	15 L min^{-1}
Auxiliary gas flow rate	1.10 L min^{-1}
Sample gas	0.97 L min^{-1}
RF power	1300 w
Runs/Passes	10/6
Sampling cone	Nickel, 1.0 mm orifice
Skimmer cone	Nickel 0.8 mm orifice
Samples per peak	20
Integration Window	80
Scan type	Escan
Detection mode	Analog
Spray chamber	Scott type (PE-Sciex)
Torch	Quartz tube

TABLE 2—*Recovery of analytes from hands of volunteers, considering collections before test shots (BS) and after test shots (AS), sample collection from around the thumb and forefinger.*

	Antimony					
	Deion. Water		HNO_3 2%		EDTA 2%	
	BS	AS	BS	AS	BS	AS
N° of shooters	32		32		32	
Max ($\mu g\ L^{-1}$)	2.73	45.6	4.52	100	6.36	162
Min ($\mu g\ L^{-1}$)	1.49	3.59	2.95	6.22	0.768	6.64
Median ($\mu g\ L^{-1}$)	2.03	12.4	3.26	14.8	2.19	62.6
Mean ($\mu g\ L^{-1}$)	2.04	13.0	3.32	20.6	2.83	68.3
SD ($\mu g\ L^{-1}$)	0.379	8.52	0.362	17.9	1.28	40.3
	Barium					
	Deion. Water		HNO_3 2%		EDTA 2%	
	BS	AS	BS	AS	BS	AS
N° of shooters	32		32		32	
Max ($\mu g\ L^{-1}$)	30.1	141	30.0	241	423	563
Min ($\mu g\ L^{-1}$)	1.86	10.5	< 0.1	23.8	10.4	41.9
Median ($\mu g\ L^{-1}$)	13.7	37.3	2.35	58.5	37.3	198
Mean ($\mu g\ L^{-1}$)	13.9	47.5	4.80	75.3	74.0	217
SD ($\mu g\ L^{-1}$)	7.25	32.0	7.12	52.2	101	139
	Lead					
	Deion. Water		HNO_3 2%		EDTA 2%	
	BS	AS	BS	AS	BS	AS
N° of shooters	32		32		32	
Max ($\mu g\ L^{-1}$)	364	870	52.6	3292	320	2895
Min ($\mu g\ L^{-1}$)	4.11	44.3	2.93	61.4	10.3	185
Median ($\mu g\ L^{-1}$)	48.6	224	9.19	165	65.3	873
Mean ($\mu g\ L^{-1}$)	93.5	263	12.6	341	117	1063
SD ($\mu g\ L^{-1}$)	98.0	186	10.3	600	153	721

TABLE 3—*Statistics of Sb, Ba and Pb concentrations ($\mu g/L^{-1}$) in samplings from the hands of shooters, before (blank) and after the shot. The sampling areas were the palm, back, thumb and forefinger palm (TF-Palm) and thumb and forefinger back (TF-Back).*

	Palm			Back			TF-Palm			TF-Back		
	Pb	Sb	Ba	Pb	Sb	Ba	Pb	Sb	Ba	Pb	Sb	Ba
Before Shot												
Max (μgL^{-1})	133	1.14	98.1	38.7	1.34	109	119	1.31	146	52.9	1.27	140
Min. (μgL^{-1})	3.43	*	2.90	*	*	*	2.76	*	2.23	2.18	*	*
Median (μgL^{-1})	21.7	0.099	18.9	12.1	0.059	18.2	29.8	0.109	17.9	13.6	0.043	16.6
Mean (μgL^{-1})	29.8	0.352	24.4	15.2	0.278	23.1	38.8	0.355	25.7	17.3	0.285	26.0
SD (μgL^{-1})	30.6	0.386	21.4	12.8	0.363	25.7	31.8	0.410	29.1	13.7	0.392	33.5
After Shot												
Max (μgL^{-1})	1206	67	236	731	81.4	482	983	68.5	299	7252	109	371
Min. (μgL^{-1})	12.5	1.25	*	1.05	0.725	*	*	4.81	*	*	1.60	*
Median (μgL^{-1})	104	6.64	11.1	41.1	3.85	37.7	162	13.8	48.6	123	15.1	66.9
Mean (μgL^{-1})	141	7.71	33.4	99.7	9.92	56.4	257	19.3	74.8	475	22.0	76.8
SD (μgL^{-1})	148	6.83	55.2	156	16.7	93.1	244	18.4	89.5	1346	24.9	89.2

* < Detection limits.

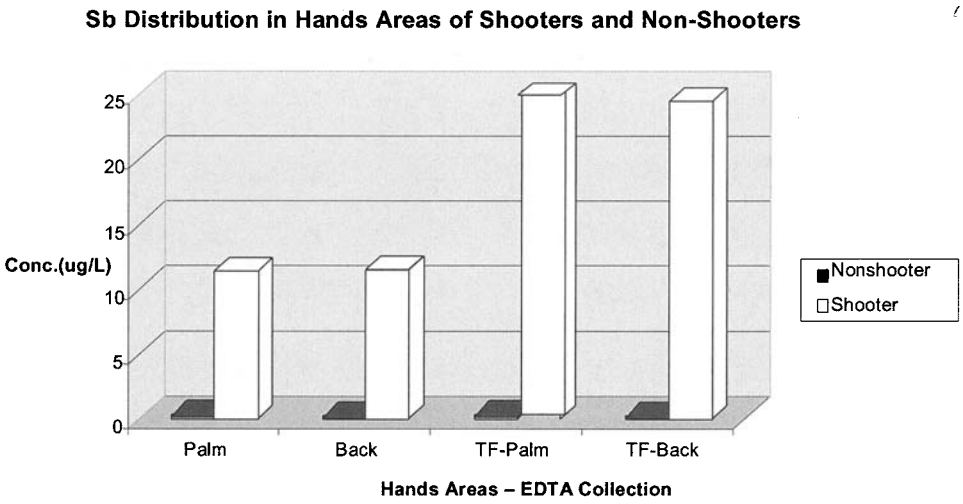

FIG. 2—*Comparison among average Sb distribution on the hands of shooters in the sampling areas studied before and after shot tests.*

bined with biological factors associated with diet and sex and age range may modify the final results.

Discussion

Considering its chemical properties, low cost and high degree of safety (compared with nitric acid) and that it does not irritate the skin, 2% EDTA solution was used for GSR sampling, prior to the determination of Sb, Ba and Pb. Stability constants of EDTA complexes for Ba and Pb in 2% solutions show its applicability to the collection method proposed, particularly since divalent cations complexes are very stable in basic or slightly acid solutions. The results showed a clear advantage of EDTA for GSR collection, allowing a good recovery of analytes, not only before but also after test shots. As expected, diluted nitric acid performed better than deionized water. Analyte measurements on extracts from some volunteers before any test shots revealed the strong influences of occupational activities and environment, but were not influenced directly by natural components present on the skin instead, since previous studies identified only potassium, sodium, magnesium, phosphorus, calcium and silicon as the main elements present (29). We also have to consider that differences observed under all firearm conditions used in this study can be decisive in influencing the recovery of GSR from hands. These arguments can be decisive in explaining random distributions in the results obtained after shot tests.

HRICP-MS provided a sensitive trace analysis of Pb, Ba and Sb in GSR extract swabs solutions, even though a quadrupole ICP-MS may be also successfully applied in this work. A comparison among quantities of analytes recovered from the hands of volunteers showed a considerable increase in the concentration of lead and antimony after the .38 firearm (of about 10 to 26 μgL^{-1}) compared with that detected on the hands before (up to 0.17 μgL^{-1}), in a way that high concentrations of antimony and lead detected together may be a strong indication of the presence of GSR, providing an effective tool for suspect identification. Differences found in GSR recoveries are related possibly to hygienic habits, occupational influences among volunteers and especially the state of preservation of the firearm, which can provide more or less GSR deposits. The collection method undertaken up to 60 min after a .38 shot may allow the identification of shooters arrested almost immediately after a shot firing. Initial results may then be related to this particular crime occurrence. The procedure related here, not only for sampling but also for GSR digestion, was simple and rapid, obviating the lack of special skills in collection and handling, so that it may be used to attend the intense demand of post-firing tests.

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