



Nanomanipulation-Coupled Nanospray Mass Spectrometry Applied to the Analysis of Trace Fibers

Journal:	<i>Journal of Forensic Sciences</i>
Manuscript ID:	draft
Manuscript Type:	Paper
Date Submitted by the Author:	n/a
Complete List of Authors:	Ledbetter, Nicole; University of North Texas, Chemistry Walton, Barbara; Central Connecticut State University, Department of Chemistry and Biochemistry Davila, Pedro; University of North Texas, Chemistry Ernest, Richard; University of North Texas, Biology Verbeck, Guido; University of North Texas, Chemistry
Keywords:	Forensic Science, Nanospray, Trace Analysis, Fiber, Micromanipulation, Gunshot residue



view

Nanomanipulation-Coupled Nanospray Mass Spectrometry Applied to the Analysis of Trace Fibers

Authors:

Nicole L. Ledbetter*, Barbara L. Walton[†], Pedro Davila*, Richard N. Ernest*, Guido F. Verbeck IV*

*University of North Texas, Department of Chemistry, PO Box 305070, Denton, TX, 76203-5070

[†]Central Connecticut State University, Department of Chemistry and Biochemistry, PO Box , New Britain, CT 06050

For Peer Review

Abstract:

This paper presents novel instrumentation of nanomanipulation coupled to nanospray mass spectrometry, which is used to directly probe trace analytes from individual fibers. New techniques are needed in order to more accurately probe trace analytes from individual fibers with analyte sensitivity and minimally evasive extraction of the sample. We are presenting new instrumentation, that has direct application to fiber analysis and trace species analysis with the capability to improve the current methods of probing. This method requires minimal sample preparation, taking the sample straight from the source to the instrument. We will demonstrate the capability of extracting a single particle of histidine, caffeine, and gunshot residue from individual fibers, and analyzing directly with nanospray MS. Nanospray MS uses small sample volumes and sample sizes, 300nl volume, making it the ideal choice to couple with the nanomanipulator. Future work will show direct cellular applications and trace forensic applications such as microphase-extraction experiments.

Keywords:

Forensic Science, Nanospray, Trace Analysis, Fiber, Micromanipulation, Gunshot residue (GSR)

Direct probing from a sample surface directly coupled to mass spectrometry is a useful tool, helping to eliminate sample preparation and analysis time. Currently there are three techniques at the forefront of direct-coupled surface sampling mass spectrometry (MS); desorption electrospray ionization (DESI)(1), surface sampling probe electrospray ionization(2), and dielectric barrier discharge ionization source (DBDI)(3). DESI sprays charged solvent droplets onto an ambient surface which ionizes neutral analytes, which are then desorbed from the surface and analyzed using MS(1,4). This technique has been used to detect trace amounts of explosives as well as sampling directly from human skin(5,6). Surface sampling probe electrospray MS uses a liquid junction between the electrospray source and the surface to dissolve and then ionize the analyte, which is then electrosprayed into the MS(2,7). This method has been

1
2
3
4 used to directly sample drugs from thin tissue slices(8) . DBDI uses a dielectric barrier discharge to create
5
6 a stable flow of plasma that desorbs and ionizes the sample off of an ambient surface then analyzes it
7
8 using MS(3). All of these techniques have great utility, but need a relatively large area, 20-100um, for
9
10 analysis.

11
12
13 Other methods have been used to analyze samples of GSR to identify the various elements which
14
15 are desired to identify and analyze. X-ray fluorescence analysis (XRF) spectrometer provides the
16
17 possibility of recording elemental mappings of samples up to 20 X 20 cm in size. Such distribution
18
19 patterns are used in GSR investigation, e.g., for shooting distance estimation(9) X-ray mapping technique
20
21 can offer a new fundamental evaluation parameter in analysis of gunshot residues with scanning electron
22
23 microscopy/energy-dispersive spectrometry, and new standards could be considered(10). Atomic
24
25 absorption spectroscopy (AAS) was used to determine the lead (Pb) pattern around bullet holes produced
26
27 by shots on test targets from the gun. Currently the European forensic laboratories use the method of
28
29 scanning electron microscopy in combination with energy dispersive X-ray microanalysis (SEM/EDX).
30
31 The investigation of gun shot residue particles in forensic laboratories is – among various chemographic
32
33 coloring methods – usually performed by SEM/EDX. The application of this technique facilitates a
34
35 concurrent analysis of both elemental composition and morphology of single particles (11).
36
37
38

39
40 As the test results can be used as evidence in court, accurate analysis of GSR is of value to
41
42 forensic scientists. The residues are principally composed of burnt and unburned particles from the
43
44 propulsive charge, as well as components from the primer, the bullet, the cartridge case and the firearm
45
46 itself(12). The detection of the inorganic and organic components of the gunpowder is not limited to a
47
48 representation of a broad list of chemicals that may be found. Besides the two energetic ingredients,
49
50 nitroglycerin and nitrocellulose, propellant powder composition includes several additives such as
51
52 gelatinizing agents, flash suppressors, plasticizers, or stabilizers(11). Time is a possible factor if any
53
54 residue sample is to be gathered from a suspect. Although GSR will be almost non-existent on a person's
55
56 hands after a few hours of normal activity, they can be retained for a longer period on hair and face swabs
57
58
59
60

1
2
3
4 and may be retained for weeks on the clothing. Typically, the airborne GSR that can be found in the
5
6 surroundings of a shooting gun is of regular or distorted spherical shape. Less frequently they form some
7
8 aggregates of spheres of various sizes and objects resembling fragments of sponge or fragments of
9
10 spherical shells.(13) If GSRs are expected on clothes, it is no longer necessary to cut out the regions of
11
12 interest, as these samples can also be investigated using a specially designed holder for clothes without
13
14 damaging the sample(9).
15
16

17
18 Micromanipulation is a significant tool in the biological and chemical sciences. It is utilized
19
20 primarily to manipulate small particles and cellular materials due to its precise movements. It is currently
21
22 being used in the biological sciences for single cell transfer(14), to isolate specific bacterial cells from a
23
24 group(15), and it has also been employed for sample preparation on MS analysis(16). Mitochondria have
25
26 been extracted from cells using micromanipulation which were then analyzed using electrophoresis(17) .
27
28 Nanomanipulation has higher resolution than micromanipulation (beyond the optical limit), allowing new
29
30 advances in the biological sciences to be made through precise movements minimizing cell damage.
31
32

33
34 There are currently no methods to probe trace analytes directly from a single fiber. The analyses
35
36 of fibers collected in the crime scene were previously checked with a low power stereomicroscope at low
37
38 magnification. Later, different techniques for further comparison were used to investigate if any of the
39
40 fibers were consistent with each other or a known source (18). One of the current methods of probing
41
42 trace analytes is the swab method. An object's surface is swabbed using a textile sampling swab that is
43
44 then put into solution to extract the analyte of interest(19). This method is not the best to collect trace
45
46 analytes, due to analyte losses and dilution of analyte concentration. Multiple steps, comes with a multiple
47
48 handling of the analyte, which can lead to sample contamination(20). Improvement in trace analyte
49
50 sampling is needed in order to be able to more accurately solve problems and collect trace evidence to
51
52 increase analyte concentration if there is not enough analyte on a single fiber utilizing the swab method.
53
54

55
56 Mass spectrometry (MS) is a useful tool, for trace analysis due to its sensitivity, specificity, and
57
58 mass accuracy enabling the analysis of many different types of compounds. Nanospray is an ideal
59
60

1
2
3
4 ionization source to couple to nanomanipulation, because it reduces sample preparation time and requires
5
6 a small amount of analyte (pmol/ul). Nanospray MS is an ionization technique that only requires a
7
8 minimum of 300 attograms (10^{-18} g) of analyte with a minimum volume of 300 μ L. Also, it is not as
9
10 affected by salts like electrospray MS is, which reduces sample preparation(21). LC-ESI-MS is used in
11
12 the trace analysis of explosives(22) and other compounds of interest; nanospray has similar mechanism to
13
14 the electrospray source except it is miniaturized and requires a lower flow rate nLs/min (23) indicating
15
16 that nanospray mass spectrometry would be a useful instrument in trace analyte analysis. These
17
18 techniques can be applied to fiber analysis, expanding the current abilities, so that trace amounts of
19
20 analyte are now able to be extracted and analyzed.
21
22
23

24 25 26 **Materials**

27
28 The solvents and chemicals utilized were Glacial Acetic Acid, Optima* LC/MS Methanol, L-
29
30 Histidine, and Caffeine (Thermo Fisher Scientific Inc.; Waltham, MA); no further purification was
31
32 necessary. Millipore water was obtained using the Milli-Q^{UF}Plus (Millipore; Billerica, MA) with better
33
34 than 18M Ω salt content. We used glass bottom dishes (0) to analyze our samples (Mat Tek Corp.;
35
36 Ashland, MA), and we probed our analyte from 100% rayon white bemberg lining. For the gunshot
37
38 residue we used four different unknown fiber samples with 1, 2, and 3 inch shot distances (Alliance
39
40 Forensics Laboratory, Ft. Worth, TX). A 6 mg sample of gun powder from an unknown source was also
41
42 provided (Alliance Forensics Laboratory, Ft. Worth, TX). We used the following instrumentation: LCQ
43
44 DECA XP Plus (Thermo Finnigan; San Jose, CA) with a nanospray ionization source (Proxeon
45
46 Biosystems; Odense, Denmark) was used to analyze the samples. L200 nanomanipulator (Zyvex;
47
48 Richardson, TX), the TE2000U Microscope (Nikon; Melville, NJ) and the PE2000b 4-Channel Pressure
49
50 injector (MicroData Instrument Inc.; S. Plainfield, NJ) were used to retrieve the analyte from the fiber.
51
52
53
54
55

56 57 **Methods**

58
59
60

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

The L-200 nanomanipulator is mounted to an inverted microscope see figure 1. The manipulator employs four positioners; two holders of end effectors either tungsten probes or microgrippers with 10nm resolution and two capillary holders with 1um resolution. A high quality piezo-motor controlled actuator nanomanipulator power supply is used in order to give precise and accurate movement in the fine and course modes of operation. The nanopositioners are controlled by a joystick, a foot pedal and computer program, which allows for precise movement. In the coarse mode of action the nanopositioners have a range of motion of 12mm in the X and Z axes and 28mm in the Y axis with. Two of the nanopositioners have a fine mode of action with a range of 100um in the Z and X axes and 10um in the Y axis. A pressure injector is used to supply up to 60psi of injection pressure and 24Hg of fill pressure to the capillaries allowing us to retrieve the analyte of interest. The nanomanipulator stage has its benefits, because it is able to hold 6-8 nanopositioners, allowing one to conduct multiple probes at a single time which increases the instrument's capabilities and effectiveness.

The Au/Pd plated nanospray tips are loaded with the solvent of interest, and then the tip is broken using the nanospray source head. A blank is run to determine any solvent contamination, and a background spectrum of the solvent is taken. The tip is then transferred to the nanomanipulator for trace analyte probing from a rayon fiber that was doped with the analyte of interest, and placed in a glass bottom dish. The rayon fiber was tacked down to ensure minimal movement of the fiber and analyte on the fiber. The particle of interest was found on the fiber, and then the nanospray tip was landed near it, less than a micrometer away. The nanospray tip injects the solvent of interest onto the analyte, which dissolves in the solvent. The solvent with the dissolved analyte is then retrieved back into the nanospray tip. The nanospray tip is then transferred directly to the nanospray ionization source and the sample analyzed. Figure 2(a) shows one of the positioners of the nanomanipulator with a nanospray tip retrieving sample. Figure 2(b) shows the tip mounted onto the nanospray assembly.

Caffeine and histidine are used to illustrate this technique. When sampling caffeine, methanol with 1% glacial acetic acid was used as the solvent, and 3ul of the solvent was loaded into the tip. The tip

1
2
3
4 was landed next to the analyte is shown in figure 3(a). The nanomanipulator used an injection pressure of
5
6 20.8psi for duration of 11msec delivered from the pressure injector, and a fill pressure of 65psi with a fill
7
8 time of 50msec. The sample is then analyzed using nanospray MS. When sampling histidine two
9
10 capillaries were used the nanospray tip filled with 3ul of 60:39:1 MeOH, H₂O, and HA the MS solvent
11
12 and another capillary filled with water seen in figure 3(b) The capillary tip injected the water onto the
13
14 histidine which dissolved in the water and then the nanospray tip retrieved the water with the dissolved
15
16 analyte. This shows the utility and flexibility of the method. The capillary was injected at 1.3psi for
17
18 100msec and the nanospray tip filled at 76.0psi for 0.5 sec repeated once. The sample is then transferred
19
20 to the nanospray MS and analyzed.
21
22
23

24 The technique was used in a similar fashion to analyze gunshot residue. Prior to analyzing the
25
26 gunshot residue from the fiber, a standard was prepared by placing a sample of gunpowder (5ug) on a
27
28 watch-glass and introduced heat to dissolve. The extraction solvent used was prepared by using a
29
30 combination of 50:50 chloroform: methanol with 1% acetic acid, due to the more nonpolar nature of the
31
32 extracted organics in the GSR. To collect the analyte from the watch glass, 6ul of the solvent was poured
33
34 using a micropipette from the rim of the watch glass. Using a new micropipette tip, the micropipette was
35
36 used to fill the analyte and transfer to a microcuvette for temporary holding. To analyze the sample, a
37
38 capillary was loaded with 5ul of the analyte and transferred to the nanospray ionization source to identify
39
40 the compounds from burning gunpowder.
41
42
43

44 The four different fabric sample swatches were labeled with the proper distance shot and from
45
46 each of the swatches a single fiber (~1mm) was pulled from the entry point using forceps. Each fiber was
47
48 observed under the microscope to identify positive identification of foreign particles. Once the particles
49
50 were located on the fiber, the nanospray tip was landed near the fiber in order to extract the particles
51
52 which is followed by injecting the analyte with the solvent. The solvent used was prepared using a
53
54 methanol and chloroform (50/50) and 1% acetic acid, which was placed in the capillary tip. The
55
56 nanospray tip injects the solvent of interest onto the analyte, which dissolves in the solvent. The solvent
57
58
59
60

1
2
3
4 with the dissolved analyte is then filled into the nanospray tip, which is then transferred directly to the
5
6 nanospray ionization source and the sample analyzed compared to the gunpowder standard.
7
8
9

10 11 **Results and Discussion**

12
13 The main purpose of this paper is to introduce this novel instrument of nanomanipulation coupled
14 to nanospray MS, as an effective tool to analyze trace fibers. Both histidine and caffeine trace particles
15 were sampled directly from a single rayon fiber using the nanomanipulator, and then directly analyzed
16 using mass spectrometry. Figure 4 shows the mass spectrum of caffeine (a) and histidine (b) after directly
17 probing trace particles from the rayon fiber. The caffeine particle had an area of $156.65\mu\text{m}^2$ calculated
18 using the Nikon software to measure the particle. The area of the histidine particle was $47.92\mu\text{m}^2$ before
19 injection took place. The limit of detection of histidine is 7pg particles when using nanospray, making
20 this an ideal method to analyze and retrieve trace analytes from fibers.
21
22
23
24
25
26
27
28
29

30
31 From the swatches containing the GSR, we were able to identify the different organic
32 components detected in the analyte extracted. We correlated eight compounds to the gun powder
33 standard. As each set of data was analyzed, the relative abundances of the organic identifiers diminished
34 based on the placement distances of the swatches. In one sample we even able to identify the inorganic
35 compounds, nickel and barium. The intensity of the inorganic compounds was low; however this was an
36 unexpected result. The metals cations may have formed salts, making them visible in the nanospray MS.
37
38
39
40
41
42
43

44 Several organic compounds were found consistent throughout each sample analyzed (Figure5).
45 The compounds that stand out in each of the samples analyzed are the plasticizers and lead. The
46 plasticizers include dibutyl phthalate, diethyl phthalate, dimethyl phthalate and lead. In the data collected
47 from the gunpowder, contact shot and one inch shot, we observed a consistency of the four previous
48 components plus ethyl centralite. All organic compounds identified showed to have similar relative
49 intensities throughout each of the data collected from the samples analyzed. However, the relative
50 intensity of dibutyl phthalate is the greatest from the data collected from the two and three inch samples.
51
52
53
54
55
56
57
58
59
60

1
2
3
4
5 Our results clearly show that the nanomanipulator coupled to nanospray MS is an effective
6 instrument to probe trace analytes from fibers. It is an improvement in trace analyte probing from a single
7 fiber, allowing for new experimental procedures to be created, and smaller amounts of analyte to be
8 sampled. The nanomanipulator reduces cost of sampling from fibers, because of the low sample
9 preparation and the reduced sample preparation time and run time. Computer control of the positioners
10 can be created to automate the procedure. Being able to extract an analyte from a single fiber allows for
11 better analysis of crime scenes, and the reduced sample size and volume required for nanospray MS gives
12 us the ability to retrieve a higher sample concentration. It is important to have a solvent to solubilize the
13 sample as well as give a steady spray flow. Some nonpolar compounds will not dissolve in any of the
14 nanospray solvents, so it is important to utilize a two capillary system with one capillary containing a
15 solvent to solubilize the analyte, and other capillary containing the nanospray friendly solvent that
16 retrieves the dissolved analyte. Some diffusion will occur and the small amount of nonpolar solvent with
17 the analyte of interest will mix with the nanospray solvent, and then can be analyzed using the MS.
18 Having the multiple nanopositioners allows us to do this. We are also capable of liquid-liquid phase
19 microextractions to sample trace analytes and gain higher sample concentration from a dilute analyte in a
20 liquid sample and then analyze it. Our next paper will illustrate our capabilities of nanomanipulating cells
21 and direct cell sampling.
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43

44 **Conclusion**

45
46 Nanomanipulation coupled to nanospray mass spectrometry is a novel technique that we
47 illustrated using caffeine, histidine and gunshot residue. We recovered trace particles of histidine and
48 caffeine from different rayon fibers, as well as gunshot residue from an unidentified fiber using the
49 nanomanipulator. We were then able to analyze the trace analyte by directly taking the analyte from the
50 source to the nanospray mass spectrometer. This clearly showed the functionality and usefulness of the
51 nanomanipulator coupled to mass spectrometry in the trace analysis of fibers, improving current methods.
52
53
54
55
56
57
58
59
60

1
2
3
4
5 Dyes and surfactants could be the reason for the unidentified ions in the GSR spectrum. Since
6
7 online pre-concentration techniques only produces positive results for metal chelates, a better option
8
9 might be a pre-concentration technique like evaporation, extraction methodologies or changing the
10
11 sampling procedure to obtain a more concentrated sample(24). However, one advantage of nanospray MS
12
13 is having the capability to analyze samples with limited volumes of up to 100 nl/ min and has limits of
14
15 detection. Future research will include direct cell probing and other trace analysis including liquid-liquid
16
17 microextraction.
18
19

20
21
22 **Acknowledgments:**

23
24 NSF-REU CHE-0648843

25
26 Zyvex Industrial Grant GN0001371

27
28 UNT Faculty Research Grant G34267

29
30 Alliance Forensics Laboratory
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51

52 **References:**

- 53
54
55 (1) RG Cooks, Z Ouyang, Z Takats, JM Wiseman. Ambient Mass Spectrometry, Science. 311 (2006)
56
57 1566-1570.
58
59
60

- 1
2
3
4
5 (2) GJ Van Berkel, AD Sanchez, JME Quirke. Thin-Layer Chromatography and Electrospray Mass
6 Spectrometry Coupled Using a Surface Sampling Probe. *Anal.Chem.* 74 (2002) 6216-6223.
7
8
9
10 (3) N Na, M Zhao, S Zhang, C Yang, X Zhang. Development of a dielectric barrier discharge ion source
11 for ambient mass spectrometry, *J.Am.Soc.Mass Spectrom.* 18 (2007) 1859-1862.
12
13
14
15 (4) JM Wiseman, BC Laughlin. Desorption electrospray ionization (DESI) mass spectrometry: a brief
16 introduction and overview. *Curr.Sep.Drug Dev.* 22 (2007) 11-14.
17
18
19
20 (5) Z Takats, I Cotte-Rodriguez, N Talaty, H Chen, RG Cooks. Direct, trace level detection of explosives
21 on ambient surfaces by desorption electrospray ionization mass spectrometry.
22
23
24 *Chem.Commun.(Cambridge, U.K.).* (2005) 1950-1952.
25
26
27
28 (6) DR Justes, N Talaty, I Cotte-Rodriguez, RG Cooks. Detection of explosives on skin using ambient
29 ionization mass spectrometry. *Chem.Commun.(Cambridge, U.K.).* (2007) 2142-2144.
30
31
32
33 (7) KG Asano, MJ Ford, BA Tomkins, GJ Van Berkel. Self-aspirating atmospheric pressure chemical
34 ionization source for direct sampling of analytes on surfaces and in liquid solutions. *Rapid*
35
36
37
38 *Commun.Mass Spectrom.* 19 (2005) 2305-2312.
39
40
41 (8) GJ Van Berkel, V Kertesz, KA Koeplinger, M Vavrek, AT Kong. Liquid microjunction surface
42 sampling probe electrospray mass spectrometry for detection of drugs and metabolites in thin tissue
43 sections, *J.Mass Spectrom.* 43 (2008) 500-508.
44
45
46
47
48 (9) Antje Berendes, Dieter Neimke, Rüdiger Schumacher and Martin Barth. A versatile technique
49 for the investigation of gunshot residue patterns on fabrics and other surfaces: m-XRF. *J. Forensic*
50
51
52
53
54
55
56
57
58
59
60 *Science* 51 (2006) 1085-1090.

- 1
2
3
4
5 (10) B Cardinetti, C Ciampini, C D'Onofrio, G Orlando, L Gravina, F Ferrari, et al. X-ray mapping
6
7 technique: a preliminary study in discriminating gunshot residue particles from aggregates of
8
9 environmental occupational origin. *Forensic Science International*, 143 (2004) 27-46.
10
11
12 (11) Désiré Laza, Bart Nys, Jan De Kinder, Andrée Kirsch-De Mesmaeker, and Cécile Moucheron.
13
14 Development of a quantitative LC-MSMS method for the analysis of common propellant powder
15
16 stabilizers in gunshot residue. *J. Forensic Science* 52 (2007) 842-850.
17
18
19 (12) Mengxia Zhao, Sichun Zhang, Chengdui Yang, Yucai Xu, Yuxiu Wen, Lianshan Sun, et al.
20
21 Desorption Electrospray Tandem MS (DESI-MSMS) Analysis of Methyl Centralite and Ethyl
22
23 Centralite as Gunshot Residues on Skin and Other Surfaces. *J Forensic Science* 53 (2008) 807-811.
24
25
26
27 (13) Zuzanna Broz'ek-Mucha. Comparison of cartridge case and airborne GSR—a study of the elemental
28
29 composition and morphology by means of SEM-EDX. *Institute of Forensic Research* 36 (2007)
30
31 398 – 407.
32
33
34
35 (14) S Yamamura, H Kishi, Y Tokimitsu, S Kondo, R Honda, SR Rao, et al. Single-Cell Microarray for
36
37 Analyzing Cellular Response. *Anal.Chem.* 77 (2005) 8050-8056.
38
39
40 (15) J Froehlich, H Koenig. Micromanipulation techniques for the isolation of single microorganisms.
41
42 *Soil Biol.* 6 (2006) 425-437.
43
44
45 (16) S Kajiyama, K Harada, E Fukusaki, A Kobayashi. Single cell-based analysis of torenia petal
46
47 pigments by a combination of ArF excimer laser micro sampling and nano-high performance liquid
48
49 chromatography (HPLC)-mass spectrometry, *J Biosci Bioeng.* 102 (2006) 575-578.
50
51
52
53 (17) H Ahmadzadeh, RD Johnson, L Thompson, EA Arriaga. Direct Sampling from Muscle Cross
54
55 Sections for Electrophoretic Analysis of Individual Mitochondria. *Anal.Chem.* 76 (2004) 315-321.
56
57
58
59
60

- 1
2
3
4 (18) Max M. Houck. Trace evidence Analysis More Cases in Mute Witnesses. Elsevier Academic
5
6
7 Amsterdam, 2004, pp 196.
8
9
10 (19) RH Bozenbury, RF Debono, LL Danylewych-May, L Fricano, L Kim, Sampling swab useful in trace
11
12 analyte detection. U.S.Pat.Appl.Publ. 2004-876478 (2005) 14.
13
14
15 (20) JBF Lloyd, RM King. One-pot processing of swabs for organic explosives and firearms residue
16
17 traces. J.Forensic Sci. 35 (1990) 956-959.
18
19
20 (21) TP White, AR Dolan, PM Bigwarfe Jr., DJ Higbee, DR Smith, TD Wood. Miniaturization of
21
22 electrospray ionization mass spectrometry. Trends Appl.Spectrosc. 4 (2002) 141-154.
23
24
25 (22) J Yinon. Analysis of explosives by LC/MS. Adv.Forensic Appl.Mass Spectrom. (2004) 231-274.
26
27
28 (23) W Henderson, JS McIndoe, Mass Spectrometry of Inorganic and Organometallic Compounds, John
29
30 Wiley & Sons, Ltd, West Sussex, England, 2005, pp. 271.
31
32
33 (24) Ernesto Morales and Alma L Revilla Vasquez. Simultaneous determination of inorganic and organic
34
35 gunshot residues by capillary electrophoresis. J. Chromotography A 1061 (2004) 225-233.
36
37
38

39 Additional Information and Reprint Requests:

40
41
42 Guido F. Verbeck, Ph.D.

43
44 University of North Texas

45
46 Department of Chemistry

47
48 P.O. Box 305070,

49
50 Denton, TX 76203

51
52 Office: 940-363-8423

53
54
55 E-mail: gverbeck@unt.edu
56
57
58
59
60

1
2
3
4
5
6
7 *Acknowledgments*

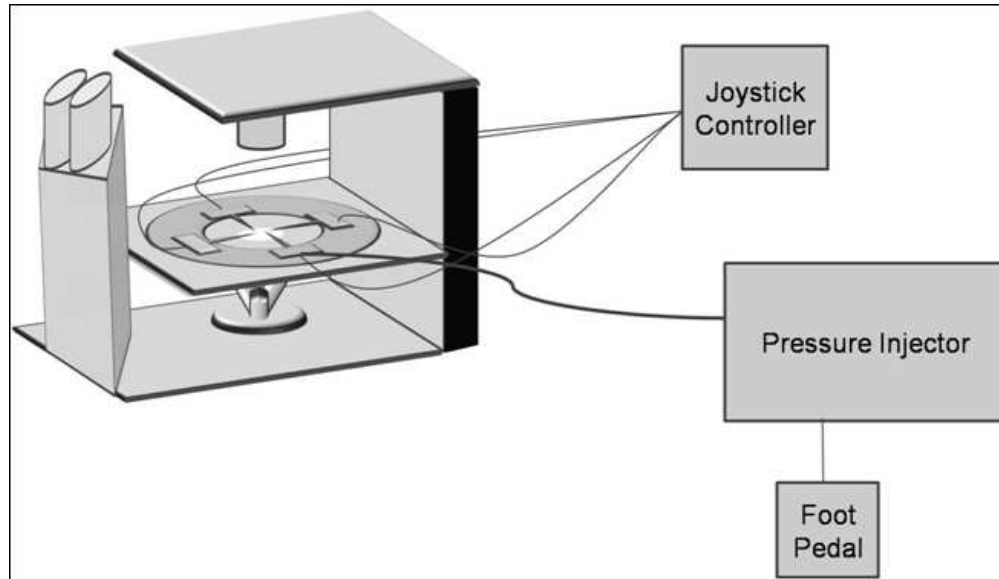
8
9 NSF-REU CHE-0648843

10
11 Zyvex Industrial Grant GN0001371

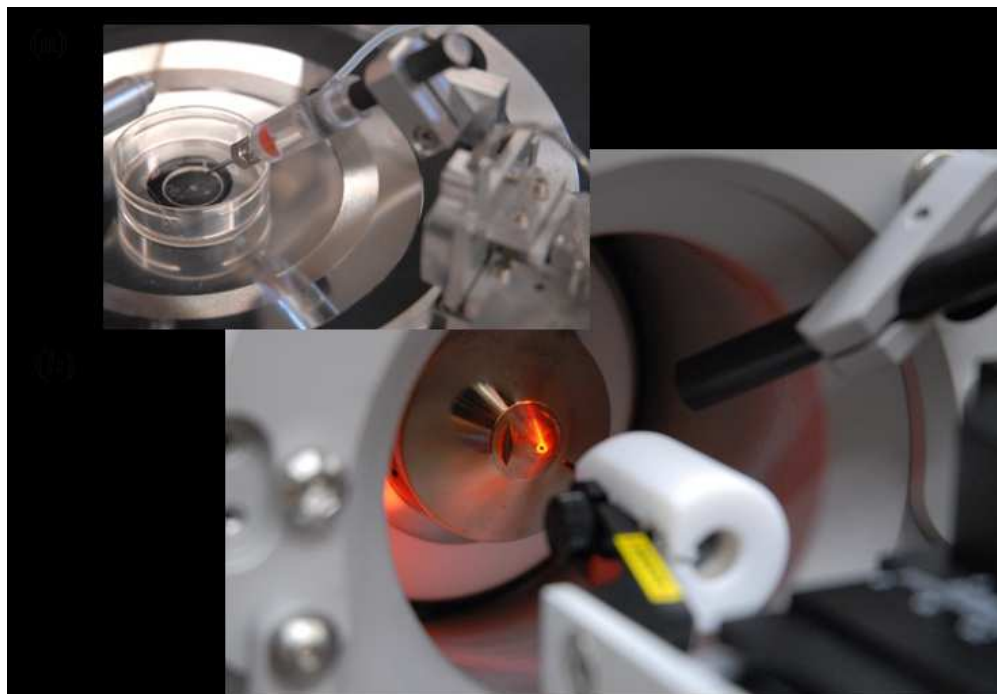
12
13 UNT Faculty Research Grant G34267

14
15 Alliance Forensics Laboratory
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

For Peer Review



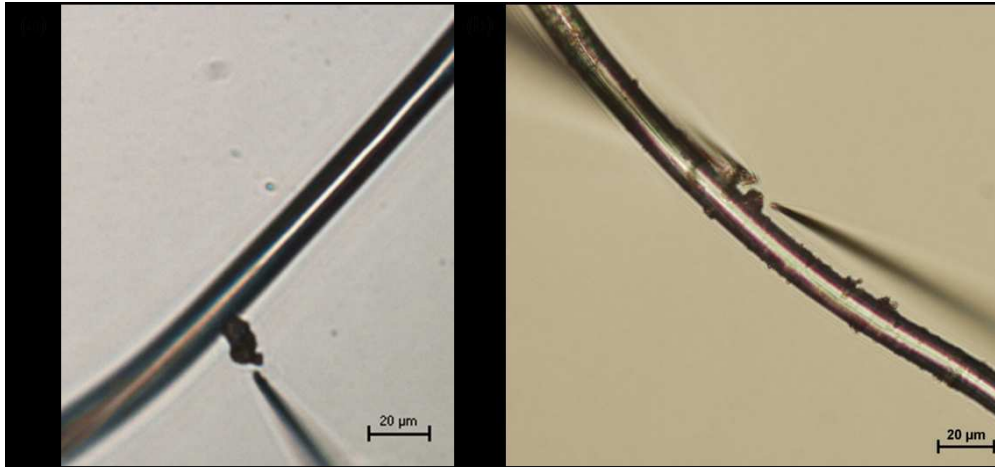
Schematic of the nanomanipulator workstation on an inverted microscope. 2 positioners are connected to a pressure injector which can be controlled through a foot pedal. The orthogonal 2 positioners are end effectors with 10 nm probing resolution. 110x63mm (150 x 150 DPI)



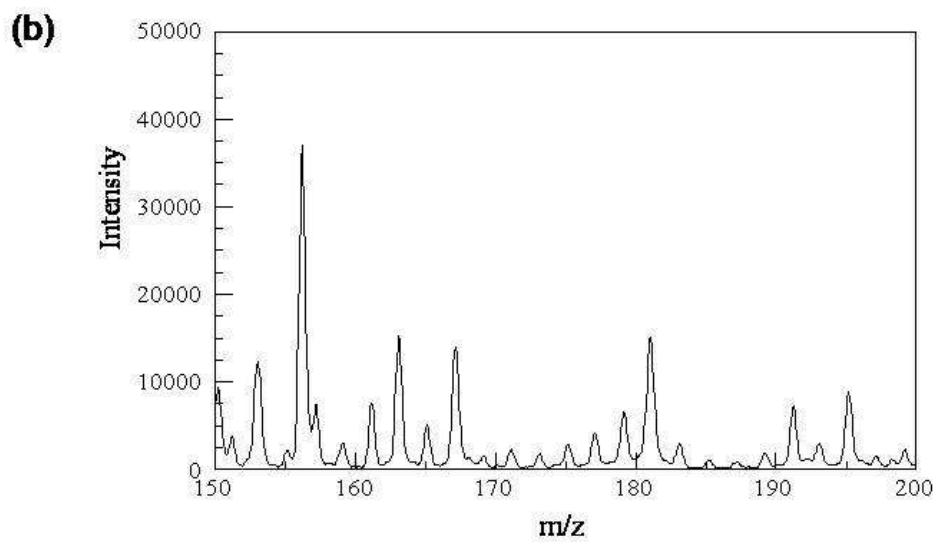
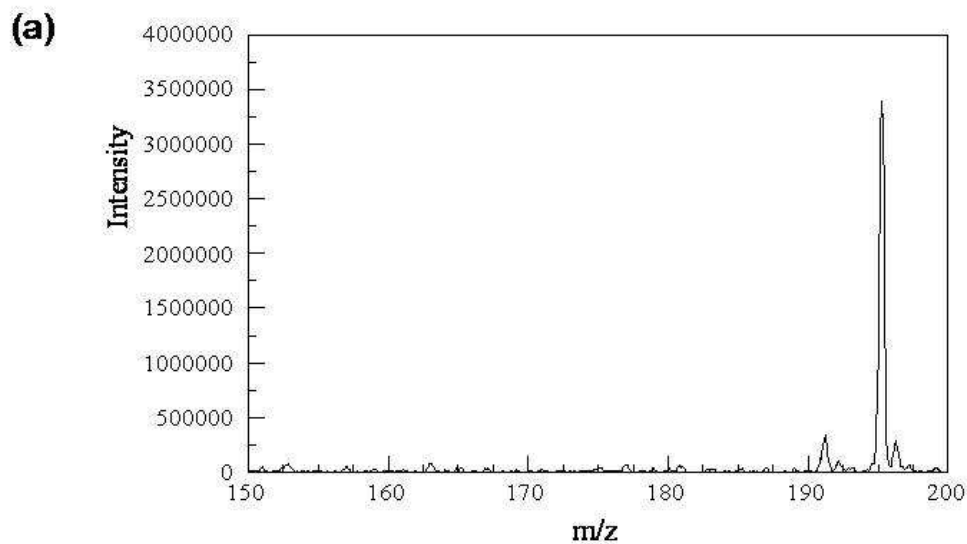
(a)The nanomanipulator positioner with the nanospray tip probing an analyte. (b) The nanospray ionization source showing the nanospray needle that was transferred directly from the nanomanipulator.

124x86mm (150 x 150 DPI)

Review

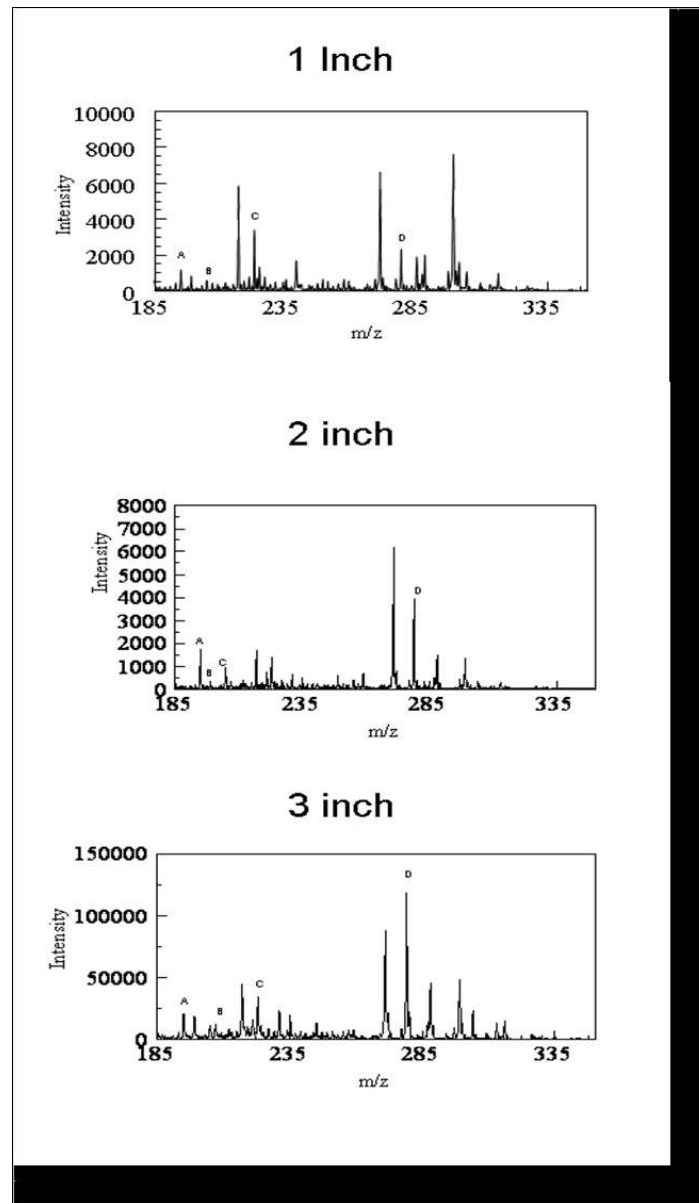


The plated rayon fiber doped with analyte before extraction. (a) The caffeine particle is on the rayon fiber and the nanospray tip is landed near the particle. (b) The histidine particle is in between the nanospray tip on the right and the capillary tip on the left.
193x89mm (150 x 150 DPI)



45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

Mass spectrum of analyte after its extraction from a single rayon Fiber (a) Caffeine, $[\text{C}_8\text{H}_{10}\text{N}_2\text{O}_2]\text{H}^+$, 195.08 m/z. (b) Histidine, $[\text{C}_6\text{H}_9\text{N}_3\text{O}_2]\text{H}^+$, 156.08m/z.
118x133mm (150 x 150 DPI)



Gunshot residue compounds identified from in each of the samples analyzed (A) Dimethyl phthalate
(B) Lead (C) Diethyl phthalate (D) Dibutyl phthalate.
111x191mm (150 x 150 DPI)

1
2
3 Figure I
45 Schematic of the nanomanipulator workstation on an inverted microscope. 2
67
8 positioners are connected to a pressure injector which can be controlled through
9
10 a foot pedal. The orthogonal 2 positioners are end effectors with 10 nm probing
11
12 resolution.
13
1415
16 Figure II
1718 (a) The nanomanipulator positioner with the nanospray tip probing an analyte. (b)
19
20 The nanospray ionization source showing the nanospray needle that was
21
22 transferred directly from the nanomanipulator.
23
2425
26 Figure III
2728 The plated rayon fiber doped with analyte before extraction. (a) The caffeine
29
30 particle is on the rayon fiber and the nanospray tip is landed near the particle. (b)
31
32 The histidine particle is in between the nanospray tip on the right and the
33
34 capillary tip on the left.
35
3637
38 Figure IV
39

40 Mass spectrum of analyte after its extraction from a single rayon Fiber (a)

41
42 Caffeine, $[C_8H_{10}N_2O_2]H^+$, 195.08 m/z . (b) Histidine, $[C_6H_9N_3O_2]H^+$, 156.08 m/z .
43
4445
46 Figure V
47

48 Gunshot residue compounds identified from in each of the samples analyzed (A)

49 Dimethyl phthalate (B) Lead (C) Diethyl phthalate (D) Dibutyl phthalate.
50
51
52
53
54
55
56
57
58
59
60