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A Review on Solid Phase Microextraction and Its Applications in Gunshot Residue Analysis

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ABSTRACT: Sample preparation is important to prepare a sample for optimum performance characteristics during analytical procedure. A review of papers on the practical applications of solid phase microextraction (SPME) is presented particularly in the analysis of gunshot residue (GSR). The general introduction on SPME and its theory are included. This review highlights the variables related to this technique during the analysis of GSR in previous studies, namely type of SPME fiber used, sampling placement, adsorption and desorption temperature and time, as well as the analytical techniques for final analysis. Apart from that, advantages and limitations of SPME based gas chromatographic methods of GSR are explored. SPME is found to be a superior sample preparation technique targeting for organic components, including GSR analysis. Gas chromatography combined with varied detection systems performed prior to SPME gives low detection limits in simpler, easier and rapid manner.

Keywords: solid phase microextraction, gunshot residue, firearm, sample preparation, gas chromatography

Introduction

In every analysis, multistep process is always involved, including sampling, preparation of sample, separation, qualitative and quantitative determination, and statistical evaluation and finally the result, decision and action. During any analytical procedure, sample preparation is the crucial step in order to prepare the sample for optimum performance characteristics on its examination. The recovery of the concentration of components in interest is aimed to increase in quantity apart from the elimination of any interfering substance that may exist in a particular sample. The efficiency and selectivity of sample preparation technique undertaken during analysis, and its applicability to the interested compounds and matrices provides benefits in numerous analytical techniques for good outcomes and results. Additionally, easy to use, inexpensive and compatible with a number of instrumental methods are the requirements of an ideal sample preparation technique. In this paper, we review the solid phase microextraction (SPME) technique and its forensic application particularly in gunshot residue (GSR) analysis.

SPME Technique

Various techniques for sample preparation have been developed previously. It was difficult to extract the total quantity of compounds by both dry and wet methods in the past for the purpose of determination and quantitation. Extraction of a sample with different solvent, evaporation steps with gas phase collection, solid phase extraction (SPE) and most recently SPME have been carried out in published studies. SPME is a sample preparation or extraction technique developed by Pawliszyn and his group in early 1990s which can be used in both in laboratory and on-site [1, 2]. This technique has gained popularity among scientists and researchers with the presence of advantages in its application in various fields, including forensic perspective.

In SPME technique, it involves the use of fused silica fiber which is very well defined in cylindrical surface geometry. This configuration allows easy assessment of analyte to and from the surface. This fiber is coated with an extracting phase. Such extracting phase can be a liquid or a solid comprised of a variety of conventional stationary phases. Different kinds of analytes, including both volatiles and non-volatiles can be extracted from different kinds of media in either liquid or gas phase. With SPME, analytes are trapped on the fiber coated with liquid polymer or solid sorbent during extraction followed by desorption into the chromatographic mobile phase for analysis [1, 2].

SPME is an equilibrium method that incorporates sampling, isolation and enrichment into a single step, applicable to gaseous, liquid and solid samples. It is a simple, convenient, reproducible and sensitive means for extraction of forensic specimen with wide linearity range and relatively good precision. SPME technique can be used for the extraction of analytes from very small samples due to its setup. Lower detection limits are permitted by its application at part per trillion concentrations. The elimination of solvents, reduction of blanks and also the extraction time to a few minutes are advantages of this extraction technique, reducing the cost and the risk of exposure to potentially hazardous solvents. As an equilibrium technique, it eliminates any exhaustive extraction of a sample. Additionally, the chemical equilibria in a particular system is not likely to be disturbed or disrupted as only a minute amounts of analytes removed on extraction. Additionally, it allows multiple sampling and preservation of the sample while minimizing the risk of contamination due to sample handling and storage [1-4].

A number of fiber types have been considered previously in SPME technique to maximize its potential applications such as polyacrylate (PA), carboxen, polydimethylsiloxane (PDMS) and divinylbenzene (DVB) as the commonly used types. Generally, the types of fiber to be considered depend on the compounds that desired to be extracted from a sample. The chemistry of compounds may decide the adsorption and desorption behaviours on a particular fiber type. Analytes varied in polarities may require different fiber chemistries. In conjunction, the thickness or area of each fiber can be differed which decides the sensitivity of the fiber. The temperature has a significant effect on the kinetics of this extraction process which depends highly on the thermal stability and volatility of the compound interested for analytical measurement. Apart from that, the optimum extraction times of this technique are affected by the combination of variables that mentioned previously [1-4].

Using SPME, there are two sampling placement of fiber for extractions that can be performed, namely direct extraction and headspace (indirect) configuration. Direct extraction mode involves the direct partitioning of analytes from the sample matrix to the extracting phase. Therefore, the coated fiber of SPME is inserted directly into the sample being examined. In latter mode, air becomes the barrier for extraction. Analytes need to be transported through the barrier to reach the coating. However, this placement of fiber protects the coating of fiber from damage by non volatile and high molecular weight interferences that could be present in sample matrix. Generally, more volatile and more hydrophilic analytes are said to be best recovered by headspace sampling. On the other hand, direct sampling from aqueous samples is the preferred mode for less volatile and more hydrophobic compounds [1, 2].

As the coated fiber placed in contact with the sample, the transport of analytes into the coating from the matrix starts. Generally, the extraction is considered to be complete when the distribution equilibrium between sample matrix and fiber coating is achieved. With the achievement of equilibrium, the amount of analytes extracted by a SPME fiber is proportional to its concentration in the sample without being dependent on the sample volume. Such relationship gives the basis for quantitation on analytes that present in sample. For both direct and headspace system, the equilibrium concentration is independent on the fiber location. Volatile analytes are extracted faster than those which are semi-volatiles due to their higher concentration in the headspace system, contributing to faster mass transport rate [1, 2].

After extraction, the fiber is withdrawn into the needle of SPME device and transferred to the injection port of instrument for desorption of analytes to be taken place. This is carried out with the piercing of the needle through a septum. Therefore, desorption of analytes are occurred thermally and transferred with the carrier gas into the column in high temperature condition. Before any analysis, it is crucial for blank runs to be performed between samples in order to ensure that the fiber and GC injection port were free of residues. Moreover, the calibration of SPME requires special care which generally consists of proper standard mixture preparation, proper calibration and establishment of the relationship between instrument response and analyte content. Finally, different detector systems can be considered for the identification and quantitative analysis, depending on the required sensitivity and selectivity of the performed analysis [1, 2].

Forensic Application in GSR Analysis

Since the development of SPME as a powerful sample preparation technique, it has been used extensively in the analysis of chemical traces in forensic applications, including toxicology [5, 6], narcotics [7-11], arson investigation [3, 12-17] and explosive identification [3, 18-23]. In this review, we review the previously published studies on the application of SPME technique in GSR analysis. The variables in such application, namely the fiber chemistry (type and thickness of stationary phase), sampling placement, adsorption and desorption temperature and time, as well as the analytical techniques for final analysis are included. Additionally, advantages of SPME in GSR analysis and its problems encountered during the analysis are also mentioned in this review.

The application of SPME technique in GSR analysis has gained popularity in recent years. In such analysis, volatile organic compounds (VOCs) from the gunpowder or propellant composition of ammunition are the interested analytes to be examined. VOCs refer to the organic compounds with significant high vapour pressure at atmospheric condition. In other words, they tend to vapourize in atmospheric pressure and temperature. Propellant, a chemical of mixture of chemicals, found inside the cartridge case is the fuel used to fire a bullet. Burning of the propellant initiated by the ignition of primer produces gases rapidly followed by the sudden increase of pressure inside the case. As a result, the bullet is detached from casing and target towards the target [24]. The formula of the gunpowder could be varied based on the use of particular ammunition due to the desired ballistics performance and stability characteristics. However, the analysis of VOCs was found to be difficult due to their low concentration in a sample. In addition, analytical standard and non-polluted sample preparation are required during their examination. Therefore, SPME is found to be the solution for these problems.

In 1998, SPME was first employed by Andrasko et al. in the examination of GSR [25]. In the performed study, the time since discharge was estimated through SPME sampling from the atmosphere inside the barrel of a shotgun. The detector systems, gas chromatography/thermal energy analyzer (GC/TEA) and gas chromatography/flame ionization detector (GC/FID) were used for its determination on the basis of the rate of escape of volatile combustion product as a function of time. With SPME technique, the analysis of GSR on the estimation of time since discharge has continued on the different types of firearms and ammunition. Following the study by Andrasko et al. (1998), the SPME technique has been used successfully for detection of various volatile compounds inside firearm barrels [26-28] as well as inside spent cartridges [29-31]. This SPME is said to be suited for headspace sampling from narrow spaces like firearm barrel by simply pushing the SPME holder inside the barrel from the muzzle of a firearm [25-27]. As compared to other headspace sampling techniques, passive adsorption process of SPME has minimized the disturbance of such extraction to the system.

For the improvement of SPME technique, modification of the liner on the injector of a chromatographic system was suggested by Andrasko and Stahling (2000) and Andrasko and Stahling (2003) [26, 27]. According to the authors, specially designed liner for desorption of VOCs was used in the injector compartment of a GC instrument. Such liner with smaller inner diameter (0.75 mm) reduces the desorption volume inside the liner and thus minimize the peak broadening of most VOCs. Apart from that, the increment in the sampling time using SPME was found to improve the sensitivity for detection of VOCs [27]. Other than TEA and FID as the detection systems as mentioned previously, mass spectrometry (MS) is another technique that was considered for GSR analysis following extraction procedure by SPME which was greatly increased in its use for forensic purposes [30-32].

Since the VOCs present in a cartridge tends to disappear into the surroundings with time, a procedure to preserve them from vaporization have to be developed for analysis. In 2003, the method of placing a shotgun shell in a vial was suggested by Wilson et al. (2003) in order to prevent the escape of VOCs before analysis [30]. In the method suggested, a hole was drilled through the center of the lid but not through the Teflon inner lining of the cap. This allowed for easy SPME sampling without opening the vial or disturbing the shell. Additionally, repetitive samplings can be performed easily without disturbing chemical equilibria in the system. There was no observable tendency for the repeatedly sampled cartridges to show lower concentration [30]. However, this was criticized by Weyermann et al. (2009) as repetitive sampling may decrease the amount of analytes with consecutive sampling, especially for smaller caliber type ammunition [31].

In 2009, SPME was adopted to extract OGSRs from a single particle of partially burnt gunpowder by Burleson et al. (2009) [33]. OGSRs from only one single particle of partially burnt gunpowder were successfully extracted by SPME and analyzed by gas chromatography/ nitrogen phosphorus detection (GC/NPD). The study showed that the new extraction method is capable of extracting trace amount of organic GSRs as signature molecules for the identification of GSR. Recently, the introduction of ion mobility spectrometry (IMS) techniques following SPME permitted the detection of trace amount of smokeless powders which are the main components in ammunition, on the discharge of any firearm. With the combination of SPME and IMS, the detection limit in nanogram range can be achieved in very short analysis time [32]. A summary of published applications of SPME on GSR analysis is presented in **Table 1**.

Table 1: Published applications of SPME on GSR analysis

Source of GSR	Sampling placement	Fiber coating	Target analyte	Extraction and desorption conditions	Limit of detection (LOD)	Final analysis	Reference
Shotgun	Headspace (exposed to atmospheric inside barrel)	PA, 85 µm	NG, 2,4-DNT, 2,6-DNT, unidentified compounds	T_C= 200^{o}C, C= 7 min, E= 30 min, T_S= r.t, T_D= 170^{o}C, D= 7 min	Peak height ca. 500 mV	GC/TEA	[25, 28]
			DPA, Nap	T_C= 200^{o}C, C= 7 min, E= 30 min, T_S= r.t, T_D= 200^{o}C, D= 7 min	Peak area 500	GC/FID	
Rifle	Headspace (exposed to atmospheric inside barrel and cartridge)	PA, 85 µm	Unidentified compounds	T_C= 200^{o}C, C= 7 min, E= 20 min, T_S= r.t, T_D= 170^{o}C, D= *	Peak height ca. 0.3-14.5 mV (muzzle), 2.0-11.8 mV (cartridge)	GC/TEA	[26]
Pistol and revolver	Headspace (exposed to atmospheric inside barrel)	Carboxen/ PDMS, 85 µm PA, 85 µm CW/DVB, *	Unidentified compounds	T_C= 170^{o}C, C= 10 min, E= 40 min, T_S= r.t, T_D= 170^{o}C, D= *	Peak height ca. 0.35-8.6 mV	GC/TEA	[27]
Spent cartridge	Headspace (exposed to atmospheric inside cartridge)	PA, 85 µm	NG, unidentified compounds	T_C= 200^{o}C, C= 7 min, E= 20 min, T_S= r.t, T_D= 170^{o}C, D= 7 min	Peak height ca. 0.5 mV	GC/TEA	[29]
			Naphthalene, biphenyl	T_C= 200^{o}C, C= 7 min, E= 20 min, T_S= r.t, T_D= 170^{o}C, D= 7 min	Peak area ca. 17-23	GC/FID	
Shotgun shell	Headspace (from corked and vial samples with shells)	PA, 85 µm	Naphthalene, biphenylene, DPA, unidentified peak (10.9 min)	T_C= 200^{o}C, C= 10 min, E= 20 min, T_S= r.t, T_D= 200^{o}C, D= 10 min	*	GC/MS	[30]
Spent cartridge	Headspace (from vial samples)	PA, 85 µm PDMS, 100 µm Carboxen/ PDMS, 75 µm	Benzonitrile, phenol, 2-ethyl-1-hexanol, naphthalene, 1,2-dicyanobenzene, DPA	T_C= 280^{o}C, C= 15 min, E= 40 min (80 min), T_S= 40^{o}C, T_D= 280^{o}C, D= 5 min	nanogram	GC/MS	[31]
Smokeless powder (Commercially available)	Headspace (from vial samples)	PDMS, 100 µm	DPA, EC, 2,4-DNT, 2-ethyl-1-hexanol	T_C= *, C= *, E= 5 – 120 min, T_S= *, T_D= 280^{o}C, D= *	0.15 – 3 ng	GC/MS	[32]
				T_C= *, C= *, E= 5 – 40 min, T_S= *, T_D= 215^{o}C, D= *, T_{DT}= 180^{o}C	0.12-1.2 ng	IMS	
Gunpowder particle	Headspace (from vial samples)	PDMS, 100 µm	DPA, NDPA, DNDPA, MC, EC	T_C= 250^{o}C, C= 5 min, E= 45 min, T_S= 95^{o}C, T_D= 250^{o}C, D= 2 min	10 ng	GC/NPD	[33]

T_C= Conditioning temperature, C= Conditioning time, E= Fiber exposition time, T_S= Sampling temperature, T_D= Desorption temperature, D= Fiber desorption time, T_{DT}= Drift tube temperature; PA= polyacrylate, PDMS= polydimethylsiloxane, CW= carbowax, DVB= divinylbenzene; NG= nitroglycerine, DNT= dinitrotoluene, DPA= diphenylamine, NDPA= nitrodiphnylamine, DNDPA= dinetrodiphenylamine, MC= methyl centralite, EC= ethyl centralite; GC= gas chromatography, TEA= thermal energy analyzer, FID= flame ionization detection, MS= mass spectrometry, NPD= nitrogen phosphorus detection, IMS= ion mobility spectrometry; *= not given

Conclusion

During analysis, the main difficulty encountered in method developments was the reliability of the sampling method. SPME was shown to be a superior alternative for such purpose, including in the analysis of GSR. In GSR analysis, desorption and analysis of volatile samples collected by SPME is most often performed using GC with different detection systems. As mentioned by numerous authors, SPME can give low detection limit with simpler, easier, faster and sensitive procedure. Considerably advantages do exist for SPME followed by sensitive analytical analysis was found to be adequate for analysis of organic volatile residues, especially in GSR.

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References

1. Arthur C. L., Pawliszyn J. (1990). Solid phase microextraction with thermal desorption using fused silica optical fibers. *Anal Chem.* 62(19): 2145-8.
2. Pawliszyn J. (1997). *Solid phase microextraction: theory and practice.* New York: John Wiley & Sons, Inc.
3. Furton K. G., Almirall J. R., Bi M., Wang J., Wu L. (2000). Application of solid-phase microextraction to the recovery of explosives and ignitable liquid residues from forensic specimens. *J Chromatog A.* 885: 419-32.
4. Namiesnik J., Zygmunt B., Jastrzebska A. (2000). Review: Application of solid-phase microextraction for determination of organic vapours in gaseous matrices. *J Chromatog A.* 885: 405-18.
5. Tranthim-Fryer D. J., Hansson R. C., Norman K. W. (2001). Headspace/solid-phase microextraction/gas chromatography-mass spectrometry: a screening technique for the recovery and identification of volatile organic compounds (VOC's) in postmortem blood and viscera samples. *J Forensic Sci.* 46(4): 934-46.
6. Kim N. Y., Park S. W. (2000). The comparison of toluene determination between headspace-solid phase microextraction and headspace methods in glue-sniffer's blood and urine samples. *J Forensic Sci.* 45(3): 702-7.
7. Coumbaros J. C., kirkbride K. P., Klass G. (1999). Application of solid-phase microextraction to the profiling of an illicit drug: manufacturing impurities in illicit 4-methoxyamphetamine. *J Forensic Sci.* 44(6) :1237-42.
8. Vu D. T. (2001). SPME/GC-MS characterization of volatiles associated with methamphetamine: toward the development of a pseudomethamphetamine training material. *J Forensic Sci.* 46(5): 1014-24.
9. Meyers J. E., Almirall J. R. (2005). Analysis of gamma-hydroxybutyric acid (GHB) in spiked water and beverage samples using solid phase microextraction (SPME) on fiber derivatization/gas chromatography-mass spectrometry (GC/MS). *J Forensic Sci.* 50(1): 31-6.
10. Brown H., Kirkbride K. P., Pigou P. E., Walker G. S. (2003). New developments in SPME, Part 1: The use of vapor-phase deprotonation and on-fiber derivatization with alkylchloroformates in the analysis of preparations containing amphetamines. *J Forensic Sci.* 48(6): 1231-8.
11. Koester C. J., Andresen B. D., Grant P. M. (2002). Optimum methamphetamine profiling with sample preparation by solid-phase microextraction. *J Forensic Sci.* 47(5): 1002-7.
12. Yoshida H., Kaneko T., Suzuki S. (2008). A solid-phase microextraction method for the detection of ignitable liquids in fire debris. J *Forensic Sci.* 53(3): .668-76.
13. Furton K. G., Almirall J. R., Wang J., Bruna J. C. (1996). A novel method for the analysis of gasoline from fire debris using headspace solid-phase microextraction. *J Forensic Sci.* 41(1): 12-22.
14. Almirall J. R., Wang J., Lothridge K., Furton K. G. (2000). The detection and analysis of ignitable liquid residues extracted from human skin using SPME/GC. *J Forensic Sci.* 45(2): 453-61.
15. Ren Q., Bertsch W. (1999). A comprehensive sample preparation scheme for accelerants in suspect arson cases. *J Forensic Sci.* 44(3): 504-15.
16. Harris A. C., Wheeler J. F. (2003). GC-MS of ignitable liquids using solvent desorbed SPME for automated analysis. *J Forensic Sci.* 48(1): 41-6.
17. Lloyd J. A., Edmiston P. L. (2003). Preferential extraction of hydrocarbons from fire debris samples by solid phase microextraction. *J Forensic Sci.* 48(1): 130-4.
18. Furton K. G., Wu L., Almirall J. R. (2000). Optimization of solid-phase microextraction (SPME) for the recovery of explosives from aqueous and post-explosion debris follwed by gas and liquid chromatographic analysis. *J Forensic Sci.* 45(4): 857-64.
19. Kirkbride K. P., Klass G., Pigou P. E. (1998). Application of solid-phase microextraction to

the recovery of organic explosives. *J Forensic Sci.* 43(1): 76-81.

20. Monteil-Rivera F., Beaulieu C., Deschamps S., Paquet L., Hawari J. (2004). Determination of explosives in environmental water samples by solid-phase microextraction-liquid chromatography. *J Chromatog A.* 1048: 213-21.
21. Muller D., Levy A., Shelef R., Abramovich-Bar S., Sonenfeld D., Tamiri T. (2004). Improved method for the detection of TATP after explosion. *J Forensic Sci.* 49(5): 935-8.
22. Psillakis E., Kalogerakis N. (2001). Solid phase microextraction versus single-drop microextraction for the analysis of nitroaromatic explosives in water samples. *J Chromatog A.* 938: 113-20.
23. Brown H., Kirkbride K. P., Pigou P. E., Walker G. S. (2004). New developments in SPME part 2: analysis of ammonium nitrate-based explosives. *J Forensic Sci.* 49(2): 215-21.
24. Meng H. H., Caddy B. (1997). Gunshot residue analysis-a review. *J Forensic Sci.* 42(4): 553-70.
25. Andrasko J., Norberg T., Stahling S. (1998). Time since discharge of shotguns. *J Forensic Sci.* 43(5): 1005-15.
26. Andrasko J., Stahling S. (2000). Time since discharge of rifles. *J Forensic Sci.* 45(6): 1250-5.
27. Andrasko J., Stahling S. (2003). Time since discharge of pistols and revolvers. *J Forensic Sci.* 48(2): 307-11.
28. Andersson C., Andrasko J. (1999). A novel application of time since the latest discharge of a shotgun in a suspect murder. *J Forensic Sci.* 44(1): 211-3.
29. Andrasko J., Stahling S. (1999). Time since discharge of spent cartridges. *J Forensic Sci.* 44(3): 487-95.
30. Wilson J. D., Tebow J. D., Moline K. W. (2003). Time since discharge of shotgun shells. *J Forensic Sci.* 48(6): 1298-301.
31. Weyermann C., Belaud V., Riva F., Romolo F. S. (2009). Analysis of organic volatile residues in 9 mm spent cartridges. *Forensic Sci Int.*186: 29-35.
32. Joshi M., Delgado Y., Guerra P., Lai H., Almirall J. R. (2009). Detection of odor signatures of smokeless powders using solid phase microextraction coupled to an ion mobility spectrometer. *Forensic Sci Int.* 188: 112-8.
33. Burleson G. L., Gonzalez B., Simons K., Yu J. C. C. (2009). Forensic analysis of a single particle of partially burnt gunpowder by solid phase micro-extraction-gas chromatography-nitrogen phophorus detector. *J Chromatog A.* 1216: 4679-83.

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