

A Novel Synchronous Fluorescence Spectroscopic Approach for the Rapid Determination of Three Polycyclic Aromatic Hydrocarbons in Tea with Simple Microwave-Assisted Pretreatment of Sample

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ABSTRACT: Many polycyclic aromatic hydrocarbons (PAHs) are carcinogenic, and some have been reported to be present in tea. People can be exposed to PAHs through tea consumption. Therefore, there is real importance for the determination of PAHs in tea. Because of the complex matrix of tea, it is hard to detect PAHs in tea without cleanup and chromatographic separation procedures. In this research, for the first time, a novel synchronous fluorescence spectroscopic approach coupling nonlinear variable-angle synchronous and matrix-isopotential synchronous scanning modes has been developed for the rapid determination of benzo(*a*)pyrene (BaP), benzo(*k*)fluoranthene (BkF), and anthracene (AN) in tea with simple microwave-assisted pretreatment of samples. This novel technique is able to resolve the spectra of the three PAHs well, even with interference from other EPA PAHs. The detection limits for BaP, BkF, and AN in tea were 0.18–0.28, 0.55–0.89, and 0.64–3.58 $\mu\text{g}/\text{kg}$, respectively, depending on various teas, with satisfactory recoveries ranging from 77.1 to 116%. The relative standard deviations achieved for BaP, BkF, and AN were 1.5, 6.6, and 8.5% for green tea; 2.9, 7.4, and 2.1% for oolong tea; and 5.6, 5.4, and 5.8% for black tea, respectively. Our results showed good correlation with those of gas chromatography–mass spectrometry. The approach developed is simple, reliable, and cost-efficient, providing an attractive alternative for the rapid selective screening of PAHs in tea.

KEYWORDS: Synchronous fluorescence spectroscopy, microwave-assisted extraction, tea, benzo(*a*)pyrene, benzo(*k*)fluoranthene, anthracene

INTRODUCTION

Tea is the traditional drink in China, and it is one of the most widely consumed drinks in the world. The consumption of tea is larger than that of coffee, cocoa drink, and carbonated drinks and is the second largest worldwide.¹ There has been a lot of research concerning the health effects of tea; for example, the theine in tea can excite the central nervous system to eliminate fatigue, and tea polyphenols can prevent arteriosclerosis and thrombosis.² However, because of both the chemical treatment of crops and the postharvest processing procedures, many residual pollutants may be present in tea^{3–5} and cause health threats to people through tea consumption.

Polycyclic aromatic hydrocarbons (PAHs) are among the most frequently reported contaminants in tea.^{6–8} PAHs have been the subject of much research because of their carcinogenicity and mutagenicity.^{9–13} Among the reported PAHs, BaP is reported to be most carcinogenic and is often considered as an important indicator of the presence of other PAHs.¹⁴ After entering the environment, PAHs can be transported long distances in the air–soil–water–plant systems and may accumulate in tea leaves and roots. PAHs have been detected in tea leaves, perhaps as a result of environmental pollution and manufacturing processes.^{5,7} The total concentration of PAHs detected in tea has been reported to be in the range of 14–2662 $\mu\text{g}/\text{kg}$ (40 samples of various types of tea)¹⁵ and 9650 ± 1200 $\mu\text{g}/\text{kg}$ (black tea samples of the same producing area).⁵ PAHs have also been detected in tea infusions: Up to 11% of PAHs in tea leaves has been reported to be transferred into the infusions, resulting in

human exposure to PAHs. Therefore, it is of great significance to determine PAHs in tea.

Until now, most of the methods used for the determination of PAHs in tea samples have been based on high-performance liquid chromatography with ultraviolet or fluorescence detection (HPLC-UV/FLD)^{1,5,8,16,17} or by gas chromatography with mass spectrometry (GC-MS).^{6,15,18} For such analyses, ultrasonic extraction^{1,5,16,17,19} or Soxhlet extraction⁶ of samples with dichloromethane or hexane was performed, and then, the extract was cleaned up on a silica and/or alumina column. Fractions eluted from the columns were further analyzed by HPLC-UV/FLD or GC-MS. The major disadvantage of these techniques is the use of large volumes of organic solvent in extraction and cleanup procedures, and they are extremely time-consuming. Moreover, the maintenance of HPLC-UV/FLD or GC-MS is complex and expensive. These disadvantages limit the development of rapid screening of PAHs in tea. Therefore, it is imperative to develop a rapid, simple, and cost-efficient technique for evaluating and screening PAHs in tea. In the present work, we propose a novel fluorescence spectroscopic approach to determine benzo(*a*)pyrene (BaP), benzo(*k*)fluoranthene (BkF), and anthracene (AN) simultaneously in tea, avoiding complicated cleanup and chromatographic separation procedures.

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Because PAHs exhibit strong native fluorescence, spectrofluorometry is especially suitable for the monitoring of PAHs, and the selectivity and sensitivity can be enhanced by synchronous fluorometric techniques, in which both excitation and emission monochromators are scanned simultaneously. Depending on the scanning rates of the two monochromators and the types of difference maintained between them, several synchronous fluorescence techniques have been developed, including constant-wavelength synchronous spectroscopy (CWSFS), constant-energy synchronous spectroscopy (CESFS), variable-angle synchronous fluorescence spectroscopy (VASFS), and matrix-isopotential synchronous fluorescence spectroscopy (MISFS).^{20–23} There has been much research reporting the applications of synchronous fluorescence approaches to the simultaneous screening of PAHs in environmental samples;^{24–28} however, to the best of our knowledge, there has been no report on the application of fluorescence spectroscopy in the determination of PAHs in tea without chromatographic separation. Instead of using a single synchronous scanning mode, VASFS and MISFS are combined in this work to provide much better flexibility and selectivity than the single technique. The sensitivity is further enhanced by coupling with first-derivative technique.

On the other hand, microwave-assisted extraction (MAE) is a technique that uses microwave energy and polar solvents to extract target compounds from matrices. MAE uses smaller volumes of solvent and is more rapid, with similar or better recoveries as compared with conventional extraction.^{29–32} There have been a few reports published in the application of MAE to the extraction of catechin, epicatechin, tea phenols, tea polyphenols, and tea caffeine from tea leaves.^{33–35} However, no report has been made of the use of MAE for the extraction of PAHs from tea leaves. We demonstrate the applicability of MAE to extract PAHs from tea samples.

The aim of this work is to provide a rapid, simple, and cost-effective technique for the rapid screening of PAHs in tea. The selectivity of the proposed approach is evaluated with standard solutions and tea extracts containing the analytes and the remaining U.S. Environmental Protection Agency (U.S. EPA) PAHs, considering both cross-interference among the three analytes and interference from the remaining EPA PAHs. The PAH amounts found in tea samples are compared with those obtained using GC-MS.

MATERIALS AND METHODS

Reagents, Standards, and Materials. The PAH standards, BaP (1 × 1 mL, ACET, 1000 mg/L, Supelco), BkF (1 × 1 mL, ACET, 1000 mg/L, Supelco), AN (98%, Sigma), and the stock 16-component PAHs mixture (methanol:methylenechloride, 1:1, Supelco) were purchased from Sigma-Aldrich (Shanghai, China) containing acenaphthene (1000 mg/L), acenaphthylene (2000 mg/L), AN (100.2 mg/L), chrysene (100.2 mg/L), benzo(*a*)anthracene (99.9 mg/L), BaP (100.1 mg/L), benzo(*b*)fluoranthene (200 mg/L), benzo(*g,h,i*)perylene (199.9 mg/L), BkF (99.9 mg/L), fluorene (200.1 mg/L), dibenz(*a,h*)anthracene (199.9 mg/L), fluoranthene (200 mg/L), indeno(1,2,3-*CD*)pyrene (100.1 mg/L), naphthalene (1000 mg/L), phenanthrene (100.1 mg/L), and pyrene (99.9 mg/L). *n*-Hexane (HPLC grade) was purchased from TEDIA Co. (United States). Water was obtained from a Milli-Q water purification system. All other chemicals were of analytical grade from Shanghai Reagent (China).

PAH Standard Solutions. Stock standard solutions of 1000 mg/L BaP, BkF, and AN in *n*-hexane were prepared and stored in a refrigerator

at 4 °C in volumetric stoppered flasks wrapped in aluminum foil to avoid degradation. The concentrations of BaP, BkF, and AN standard solutions were diluted to 1.00, 1.00, and 4.00 mg/L by *n*-hexane, respectively.

Samples. Tea samples were purchased from markets in Xiamen. Four green tea samples were produced in Sichuan province, Hu'nan province, An'hui province, and Fujian province, respectively. Black tea and chrysanthemum tea were produced in An'hui province. Other tea samples were produced in Fujian province (China).

Sample Preparation. Samples were extracted using a domestic microwave oven, Galanz (G7020 II YSL-V1). The MAE process was carried out at the irradiation power of 140 W. Satisfactory recoveries of BaP were obtained at this irradiation power because of the high efficiency of the microwave extraction. Dried tea samples were homogenized, and 0.5–1.0 g of tea was placed in an uncovered conical flask (100 mL) and then extracted by microwave with 15 mL of dimethyl sulfoxide (DMSO) for 4 min. Meanwhile, a cup of 400 mL of water was placed in the microwave oven to avoid the sample overheating. The number of samples extracted per run is four. After cooling, the extract was filtered through glass fiber into a separating funnel. The extract was then mixed with 35 mL of water containing 2% sodium sulfate and twice extracted with 15 mL of *n*-hexane.³⁶ After extraction, solvent was removed with a rotary evaporator, and the concentrated extract diluted to 5 mL by DMSO.

Analysis of PAHs. All fluorescence spectra were obtained with a laboratory-constructed, computer-controlled MYF spectrofluorimeter³⁷ equipped with a 150 W xenon arc lamp and slit band passes set at 5 nm for excitation and emission monochromators. Both monochromators were controlled by a personal computer through a software package written in Turbo C 2.0. Experimental data were collected by the computer via the software package. A program written in Visual Basic for Excel was used for further data processing. Theoretical three-dimensional spectra calculated from the excitation and emission spectral data were used to acquire a suitable scanning route by means of this program. Synchronous fluorescence spectra were recorded with the selected scanning route. Derivative spectra were then obtained by the electronic differential system of the spectrofluorimeter. Calibration curves were made by additions of BaP, BkF, and AN to tea extracts. A quartz cuvette with a path length of 1 cm was used throughout.

Method Validation. For validation, recovery tests were carried out as followed. Five tea samples were spiked with six dilutions of a standard mixture of BaP, BkF, and AN. The concentrations of BaP, BkF, and AN ranged from 0 to 60 µg per kilogram dried tea. These standard solutions were added in tea before extraction. The concentrations of BaP, BkF, and AN were determined. Finally, the new method was compared with results using GC-MS.

Conditions for Comparison Experiment. GC-MS measurements were performed on a Shimadzu GCMS-QP2010 (Japan) with DB-5MS capillary column (30 m × 0.25 mm × 0.25 µm) (Agilent). The injector temperature was 250 °C; the injection volume was 2 µL (splitless). Helium was the carrier gas at a flow rate of 1.2 mL/min. The column temperature was programmed to start at 100 °C for 2 min and then increased to 180 °C at a rate of 10 °C/min and held for 5 min. After that, the temperature was increased at a rate of 15 °C/min up to 280 °C and held for 10 min. Mass spectrometer conditions were as follows: electron impact ionization energy, 70 eV; ion source and transfer line temperatures, 250 and 280 °C, respectively. Selected ion monitoring (SIM) mode was used for identification. The SIM ions (*m/z*) selected for quantification are 178 for AN and 252 for BaP as well as for BkF.

RESULTS AND DISCUSSION

Optimization of Scanning Routes. To produce the best scan spectrum (highest signal values, interference-free bands) in

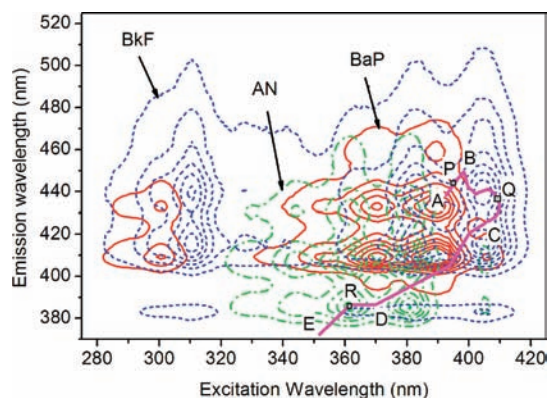


Figure 1. Theoretical three-dimensional spectra of BaP ($5 \mu\text{g/L}$, solid lines), BkF ($5 \mu\text{g/L}$, dashed lines), and AN ($10 \mu\text{g/L}$, dash-dotted lines) in DMSO. The bold folded line ABCDE is the selected NLVA-MISF scanning route. Folded lines AB, BC, and DE are the detection routes of BaP, BkF, and AN, respectively.

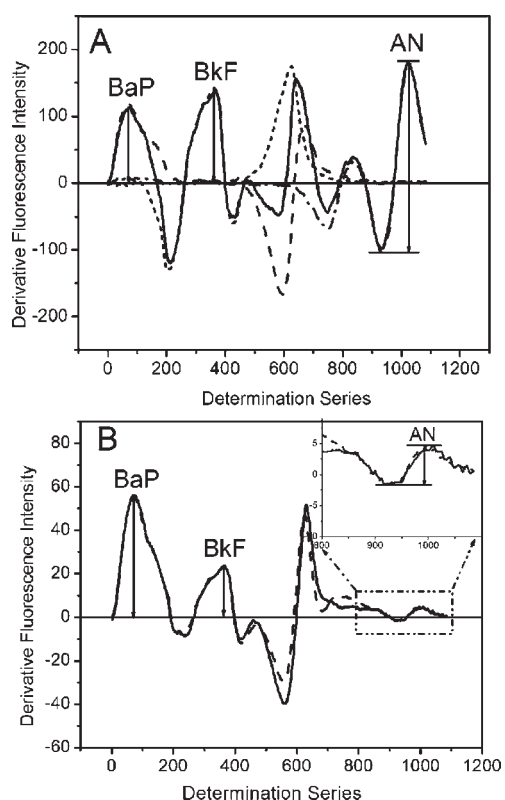


Figure 2. (A) First-derivative NLVA-MISF spectra of BaP ($1 \mu\text{g/L}$, dashed curve), BkF ($3 \mu\text{g/L}$, dotted curve), AN ($16 \mu\text{g/L}$, dash-dotted curve), and a mixture of the three PAHs at the same concentration as above. (B) First-derivative NLVA-MISF spectra of a mixture of EPA 16 PAHs (dashed curve, the concentrations of BaP, BkF, and AN among the 16 PAHs were $5 \mu\text{g/L}$, respectively) and a mixture of BaP, BkF, and AN (solid curve, concentrations the same as those in EPA 16 PAHs).

derivative nonlinear variable-angle coupled MISFS (DNLVA-MISFS), it is necessary to select an optimum scan route. After recording the excitation and emission fluorescence spectra of BaP, BkF, and AN, we calculated the theoretical three-dimensional spectra from a pair of excitation and emission spectra by use of a homemade data-processing program. BaP, BkF, and AN

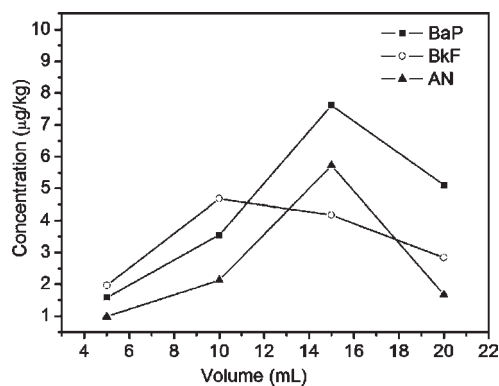


Figure 3. Effect of the volume of solvent on the extraction of BaP (■), BkF (○), and AN (▲) from tea.

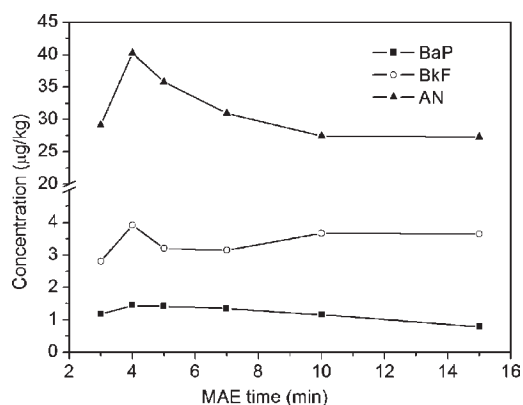


Figure 4. Effect of MAE time on the extraction of BaP (■), BkF (○), and AN (▲) from tea.

are structurally similar and exhibit seriously overlapped spectra, as seen in Figure 1. The fluorescence spectrum of BaP was affected by the spectra of BkF and AN together. Considering signal values, interference, and scattering lights, we chose point P ($\lambda_{\text{ex}} = 395 \text{ nm}$, $\lambda_{\text{em}} = 444 \text{ nm}$) as the detection point of BaP, avoiding interference from BkF and AN, and point R ($\lambda_{\text{ex}} = 361 \text{ nm}$, $\lambda_{\text{em}} = 386 \text{ nm}$) as the detection point of AN avoiding interference from BaP and BkF. For the detection of BkF, anticipating BaP as the interference, point Q ($\lambda_{\text{ex}} = 409 \text{ nm}$, $\lambda_{\text{em}} = 437 \text{ nm}$) was chosen. We then searched for the fluorescence intensity of BaP at this point and obtained a set of λ_{ex} and λ_{em} with the same intensity, which gave a trajectory for detection of BkF. Other points could be selected to make up a whole scanning route ABCDE.^{23,38} Thus, the individual signals of BaP, BkF, and AN were obtained by scanning along route AB, route BC, and route DE, respectively.

DNLVA-MISFS Measurements. Scanning along the selected trajectory, we recorded NLVA-MISF spectra of BaP, BkF, and AN, and peaks for BaP, BkF, and AN appeared in turn. The selectivity was further enhanced by combining the first-derivative technique. The scanning time for a first-derivative NLVA-MISF spectrum is only about 50 s. For convenient observation and quantitative measurements, NLVA-MISF spectra were plotted with determination series as x -axis and the derivative fluorescence intensity as y -axis, as indicated in Figure 2A. The positive peaks were measured for the detection of BaP and BkF, and the positive- and negative-derivative peak was measured for the

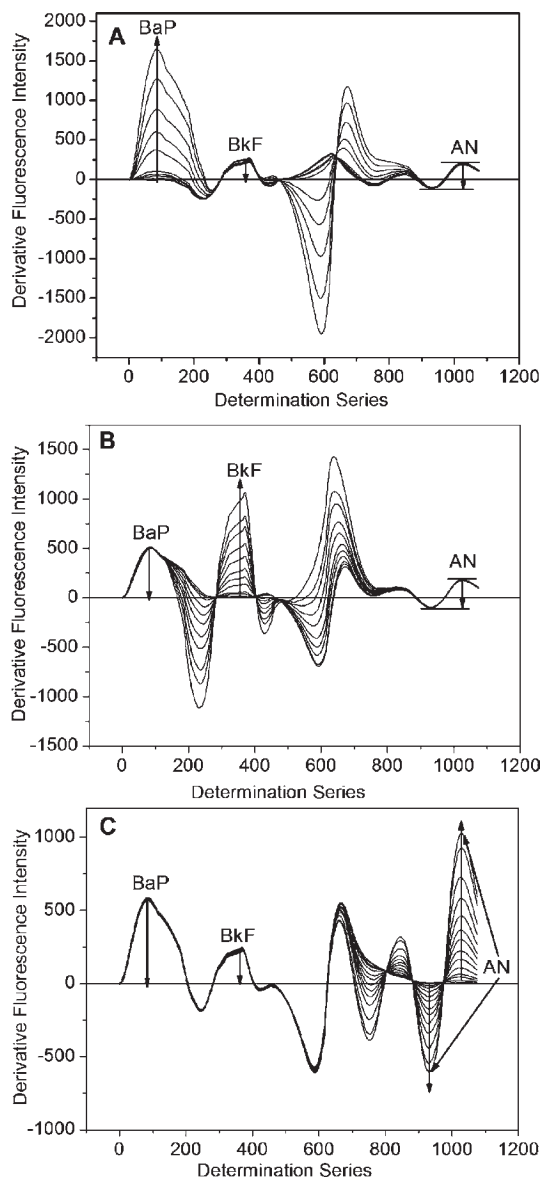


Figure 5. First-derivative NLVA-MISF spectra of mixtures of BaP, BkF, and AN at different concentrations. (A) The concentration of BaP is 0.4–50 nmol/L in the presence of 20 nmol/L BkF and 90 nmol/L AN. (B) The concentration of BkF is 0.8–100 nmol/L in the presence of 20 nmol/L BaP and 90 nmol/L AN. (C) The concentration of AN is 5.5–580 nmol/L in the presence of BaP and BkF at 20 nmol/L, respectively.

detection of AN. Figure 2B shows the first-derivative NLVA-MISF spectra of a mixture of BaP, BkF, and AN and the standard solution of EPA 16 PAHs mixture. It is obvious that there is neither cross-interference among BaP, BkF, and AN nor interference from the remaining EPA PAHs on the analytes.

Optimizing the Parameters of MAE. Usually, the extraction efficiency could be influenced by several extraction conditions such as the solvent and its volume and extraction time. These parameters were optimized to obtain best extraction performance.

Solvent Selection. Several extraction solvents used frequently in the tea studies have been investigated, including methanolic potassium hydroxide (KOH–MeOH) solution, acetone, dichloromethane (DCM), and DMSO. The results indicate that

Table 1. Detection Limits for BaP, BkF, and AN in Various Teas

tea	detection limit ($\mu\text{g}/\text{kg}$)		
	BaP	BkF	AN
green tea	0.28	0.89	3.58
oolong tea	0.26	0.66	0.75
black tea	0.18	0.55	0.64

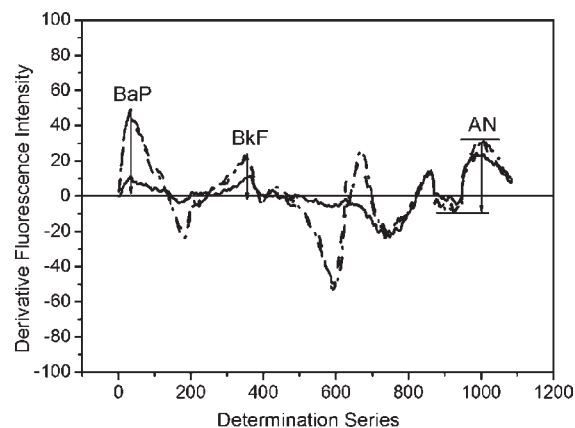


Figure 6. First-derivative NLVA-MISF spectra of tea sample (solid curve), tea sample spiked with a mixture of EPA 16 PAHs (dashed curve, the concentrations of BaP, BkF, and AN among the 16 PAHs were 8.0 $\mu\text{g}/\text{L}$, respectively), and spiked with a mixture of BaP, BkF, and AN standard solutions (dash dotted curve, concentrations were the same as those in EPA 16 PAHs).

the fluorescence intensity of BaP extracted in DMSO is strongest. The extraction efficiency of acetone or acetone/DCM is not high. This is mainly due to the easy volatilization of the solvents. Moreover, DMSO is an important polar solvent with lower toxicity than the above solvents and is preferable to suppress matrix interference.³⁹ It was therefore chosen as the extraction solvent.

Effect of Solvent Volume and MAE Time. In the MAE process, the amount of solvent can affect the solution temperature and recovery significantly, bringing the positive and negative influences to the extraction efficiency with increasing amount of solvent.³³ In our trials, the volume of DMSO used in extractions was optimized. As shown in Figure 3, the extraction of the three PAHs increased at first and then decreased when the solvent was more than 15 mL. Because DMSO is polar and the microwave extraction is very quick, microwave energy will transfer fast, resulting in the easy extraction of PAHs.^{30,32}

Finally, the MAE time was explored from 3 to 15 min with the volume of DMSO at 15 mL. The three PAHs showed the similar behavior, with maximum extraction being attained at 4 min (Figure 4). If MAE time was longer than 4 min, the extraction of AN was decreased with the increase of time. Therefore, the MAE time of 4 min was used in the following experiments.

Analytical Features of the Proposed Method. With the optimum scanning route, the first-derivative NLVA-MISF spectra of BaP, BkF, and AN were resolved well. With concentration of BaP up to 50 nmol/L, spectra of BkF and AN were generally invariable. Similarly, with concentrations of BkF and AN up to 100 and 580 nmol/L, there was no significant interference on the other two analytes (Figure 5).

Table 2. Recoveries of PAHs in Tea Samples of the DNLVA-MISFS Method

no spiking ($\mu\text{g}/\text{kg}$)			spiked amount ($\mu\text{g}/\text{kg}$)			detected amount ($\mu\text{g}/\text{kg}$)			recovery (%)		
BaP	BkF	AN	BaP	BkF	AN	BaP	BkF	AN	BaP	BkF	AN
4.60	ND ^a	64.3	0.00	10.0	60.0	4.70	8.60	123	NA ^b	86.0	98.2
13.2	9.85	50.8	10.0	20.0	40.0	13.8	18.3	97.1	92.0	91.5	82.0
4.65	10.2	87.9	20.0	30.0	0.00	31.2	36.1	50.6	89.8	87.3	NA
21.6	15.3	48.9	30.0	0.00	20.0	29.5	11.3	111	82.7	NA	116
12.8	21.6	130	5.00	8.00	12.0	25.9	22.6	58.1	86.0	90.6	77.1
			25.0	15.0	8.00	34.0	38.0	137	84.6	109	82.5

average recoveries \pm SDs (%) 87.1 \pm 3.8 93.1 \pm 9.4 90.9 \pm 16

^a Not detectable. ^b Not applicable.

Table 3. Comparison of the Results Obtained by DNLVA-MISFS and by GC-MS

no.	BaP ($\mu\text{g}/\text{kg}$)		BkF ($\mu\text{g}/\text{kg}$)		AN ($\mu\text{g}/\text{kg}$)	
	DNLVA-	GC-MS	DNLVA-	GC-MS	DNLVA-	GC-MS
	MISFS		MISFS		MISFS	
1	18.7	26.1	24.0	33.1	52.8	70.8
2	17.3	17.4	43.1	46.0	90.6	98.7
3	8.18	8.39	ND	ND	37.9	34.1
4	8.44	7.13	6.44	6.09	34.0	37.2
5	0.28	ND ^a	10.4	8.73	4.69	5.41
6	11.9	13.3	10.5	9.20	7.69	5.40

^a Not detectable.

Detection Limits. The limit of detection (LOD) was evaluated on the basis of three times the standard deviation (SD) on the (nearly) blank samples (LOD = 3 SD/bk, $n = 9$). The LODs for BaP, BkF, and AN in green tea, oolong tea, and black tea are shown in Table 1. These three teas are representative of nonfermented, semifermented, and fully fermented teas, respectively.

Precision. The precision of the method was calculated as repeatability by using the relative standard deviation (RSD) for replicate detections of five tea samples from the same source. The RSDs achieved for BaP, BkF, and AN were 1.5, 6.6, and 8.5% for green tea; 2.9, 7.4, and 2.1% for oolong tea; and 5.6, 5.4, and 5.8% for black tea, respectively.

Interference Investigation on Real Samples. Figure 6 shows the first-derivative NLVA-MISF spectra of a tea sample and tea sample spiking with a mixture of EPA 16 PAHs and with the mixture of BaP, BkF, and AN standard solutions. It can be seen that the peaks of BaP, BkF, and AN appear in sequence, and the peak positions and intensities of each PAHs in the two spectra are coincident, indicating that there is no interference from the remaining EPA PAHs on the spectra of BaP, BkF, and AN in tea samples.

Method Validation. Recoveries from Spiked Tea Samples. To investigate the accuracy and the reliability of the proposed method, the recoveries of BaP, BkF, and AN were studied on five tea samples spiked with six concentration levels of BaP, BkF, and AN standard solutions. These standard solutions were added to the tea samples before the MAE. The recoveries ranged from 77.1 to 116%, as indicated in Table 2.

Comparison with GC-MS. The proposed method was also compared with GC-MS. The sample extractions were same as described in the Materials and Methods, followed by a cleanup step

Table 4. PAHs Levels ($\mu\text{g}/\text{kg}$) in 15 Tea Samples

samples	BaP	BkF	AN
green tea A	23.0 \pm 2.1	34.8 \pm 4.6	348 \pm 14
green tea B	11.2 \pm 1.6	17.4 \pm 5.3	131 \pm 9.5
green tea C	ND ^b	37.3 \pm 8.5	ND
green tea D	4.54 \pm 0.1	26.9 \pm 2.5	190 \pm 11.2
oolong tea A	7.55 \pm 1.2	ND	33.2 \pm 3.6
oolong tea B	67.7 \pm 4.3	ND	330 \pm 35
oolong tea C	26.9 \pm 1.6	79.2 \pm 11	ND
oolong tea D	39.4 \pm 3.5	ND	27.9 \pm 1.4
black tea	9.40 \pm 0.9	ND	132 \pm 5.6
pu'er tea A	5.26 \pm 0.2	26.0 \pm 2.1	44.8 \pm 2.2
pu'er tea B	22.2 \pm 1.7	166 \pm 8.2	201 \pm 12
pu'er tea C	9.30 \pm 0.3	20.3 \pm 1.2	175 \pm 8.8
chrysanthemum tea A	ND	41.6 \pm 5.9	40.5 \pm 5.6
chrysanthemum tea B	2.59 \pm 0.1	53.2 \pm 6.1	55.5 \pm 10.6
chrysanthemum tea C	ND	16.6 \pm 1.3	ND

^a Mean \pm standard deviation of duplicate analyses. ^b Not detectable.

according to the literature.³⁶ Briefly, the concentrated extracts were mixed with a triple volume of water and then loaded into a preactive C18 column. The column was washed with 15 mL of acetonitrile/water (1/4, v/v) to elute interfering material, then eluted with 10 mL of *n*-hexane to collect PAHs. The *n*-hexane elute was concentrated by rotary evaporation to 1 mL, and an aliquot of 2 μL was injected into GC-MS analysis. The results are shown in Table 3. We applied the *t* test to evaluate the difference between the proposed method and the GC-MS method. Take BaP, for example; when the confidence interval (*P*) is 0.95, the absolute value of calculated *t* is 0.28, which is smaller than $t_{0.05}(10) = 2.23$, showing there is no significant difference between these two methods. The absolute values of calculated *t* for BkF and AN in the comparison experiments are 0.15 and 0.20, respectively. Therefore, there is no significant difference between the results obtained by the proposed method and those by the GC-MS method.

Applications. The proposed method was applied to rapid screening of PAHs in 15 tea samples, including nonfermented tea (green tea, chrysanthemum tea), semifermented tea (oolong tea), and fully fermented tea (black tea and pu'er tea). As shown in Table 4, the detected concentration of BaP, BkF, and AN was from ND to 348 $\mu\text{g}/\text{kg}$. However, there was no obvious trend of the contaminated BaP, BkF, and AN in tea from various areas in China or in different kinds of tea.

In conclusion, the combination of a matrix-isopotential synchronous fluorescence technique with variable-angle synchronous fluorescence technique is a novel approach for the rapid selective determination of PAHs in tea samples free from tedious cleanup procedures. MAE has been introduced in the efficient extraction of PAHs in tea. The results indicated that the approach developed is able to resolve the spectra of BaP, BkF, and AN well, even tolerating the interference from remaining EPA PAHs. The results obtained by DNLVA-MISFS were in good agreement with those obtained by GC-MS. This novel technique is simple, reliable, and cost-efficient and provides an attractive alternative for rapid selective screening of PAHs in the safety evaluation of tea samples.

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Notes

Safety

Because of the high toxicity of PAHs, special attention was taken during all experiments. All compounds were handled in a fume hood and appropriate protective safety glasses, gloves, and lab coats were used.

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ABBREVIATIONS USED

PAHs, polycyclic aromatic hydrocarbons; U.S. EPA, U.S. Environmental Protection Agency; BaP, benzo(a)pyrene; BkF, benzo(k)fluoranthene; AN, anthracene; CWSFS, constant-wavelength synchronous spectroscopy; CESFS, constant-energy synchronous spectroscopy; VASFS, variable-angle synchronous fluorescence spectroscopy; MISFS, matrix-isopotential synchronous fluorescence spectroscopy; MAE, microwave-assisted extraction; NLVA-MISFS, nonlinear variable-angle coupling matrix-isopotential synchronous fluorescence spectroscopy; KOH–MeOH, methanolic potassium hydroxide; DMSO, dimethyl sulfoxide; DCM, dichloromethane; HPLC-UV/FLD, high-performance liquid chromatography with ultraviolet or fluorescence detection; GC-MS, gas chromatography coupled with mass spectrometry; LOD, limit of detection; RSD, relative standard deviation

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