Development of octadecyl-functionalized-nanotubular TiO₂/Ti wire solid-phase microextraction fiber†

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An octadecyl-functionalized solid-phase microextraction (SPME) fiber was prepared by sol–gel technology with an anodized Ti wire as the substrate and dimethylloctadecyl[3-(trimethoxysilyl)propyl]ammonium chloride (C₁₈-TMS) and tetraethoxysilane (TEOS) as sol–gel precursors. The anodized Ti wire has high mechanical strength and has numerous titanol groups on its surface for sol–gel reactions, consequently offering better performances than the commercial fragile fused silica substrate. The C₁₈-TMS/TEOS sol–gel coated fiber has good thermal stability and stability against organic solvents. There is no loss in the performance after 100 cycles of exposure to acetonitrile and high temperature (300 °C) in turn. The developed fiber with a very thin (ca. 2 μm) coating thickness exhibits comparable or even superior extraction capability relative to the commercial 100 μm polydimethylsiloxane (PDMS) fiber. Under optimized conditions, the detection limits for the polycyclic aromatic hydrocarbons are in the range of 0.003 to 0.025 μg L⁻¹ with a linear range from 0.01 to 20 μg L⁻¹. The proposed method was successfully applied in the analysis of environmental water samples with the recoveries ranging from 85.3 to 101.8%.

1 Introduction

In recent years, solid-phase microextraction (SPME) has been widely applied as a simple, rapid, and solvent-free sample preparation technique.¹–⁴ It integrates sampling, concentration, and introduction into one step, and has been coupled with gas chromatography (GC)⁵–⁷ and high performance liquid chromatography (HPLC).⁸–¹⁰ SPME is a non-exhausted sample preparation technique and is based on the distribution of analytes between a sample matrix and a sorbent coating immobilized on a fiber substrate. Therefore, preparation and selection of a suitable sorbent coating is extremely important for the SPME approach to achieve high sensitivity and selectivity. To date, commercially available SPME fibers with a variety of coatings such as polydimethylsiloxane (PDMS),¹¹,¹² polyacrylate (PA),¹³ carbowax (CW),¹⁴ polydimethylsiloxane–divinylbenzene (PDMS–DVB)¹⁵ and carboxen–polydimethylsiloxane (CAR–PDMS)¹⁶ have been widely applied in the analysis of trace organic compounds in many areas. Meanwhile, a lot of novel materials such as carbon nanotubes,¹⁷ graphene¹⁸,¹⁹ and metal-organic frameworks²⁰ have also been exploited as coatings for home-made SPME fibers to meet the increasing demands of trace analysis.

However, most of these fibers do have some drawbacks: polymeric coatings usually exhibit relatively low operating temperatures, organic solvent instability and less selectivity; some new type inorganic coatings lack efficient binding to the fiber substrate.

Utilizing sol–gel technology can overcome some of these problems²¹,²² because the sol–gel chemistry can provide an efficient way to combine the coating material with the fused silica substrate through covalent bonding. Various coating materials including porous materials,²³ nanomaterials,²⁴,²⁵ and ionic liquids²⁶,²⁷ have been introduced in SPME for analysis of different kinds of compounds via sol–gel technology. In order to be incorporated into the sol–gel network through chemical bonds, the coating material often needs to be functionalized so that it can react with the sol–gel components. As the functionalization is usually a complicated and time-consuming synthesis process, it is more convenient and efficient to directly use functionalized alkoxysilane precursors to prepare sol–gel coated fibers. A few fibers were prepared using this approach. Gbatu et al.⁸ developed an octyl-functionalized fiber using methyltrimethoxsilane and n-octyltriethoxysilane as the sol–gel precursors for determination of organometals. Azenha et al.²⁸ reported a phenyl-functionalized SPME fiber coating obtained from methyltrimethoxsilane and phenyltrimethoxsilane.

Most commercial and home-made SPME fibers use the fragile fused silica rod as the substrate, which greatly limits the application of SPME due to the low mechanical strength of
the substrate. Applying high mechanical strength metallic wires to replace the fused silica rods can overcome this problem.\textsuperscript{29-31} Among them, Ti wire has attracted a lot of attention due to its corrosion resistance, high stability and biocompatibility.\textsuperscript{32,33} Moreover, an anodized Ti wire on which perpendicularly oriented and well-aligned TiO$_2$ nanotube arrays are formed would be an interesting substrate for SPME because it has ultrahigh specific surface area and is easy to functionalize.

In this study, a new octadecyl-functionalized SPME fiber coating was prepared by sol–gel technology for the analysis of polycyclic aromatic hydrocarbons (PAHs) combined with HPLC analysis. An anodized Ti wire was used as the substrate providing high mechanical strength. The nanotubular surface provides abundant surface titanol groups for sol–gel reactions.

2 Experimental

2.1 Chemicals and reagents

Titanium wire (Φ 0.20 mm, 99.9% in purity) was purchased from Alfa Aesar (MA, USA). Hydrofluoric acid (40.0%) was purchased from Tianjin Fuyu Fine Chemicals Co., Ltd (Tianjin, China). Tetraethoxysilane (TEOS) was purchased from Acros Organics (NJ, USA). Dimethyloctadecyl[3-(trimethoxysilyl)propyl]ammonium chloride (C$_{18}$-TMS) and trifluoroacetic acid (TFA) were purchased from Aladdin Reagent (Shanghai, China). n-Hexane, acetone and dichloromethane were purchased from Sinopharm Chemical Reagent (Shanghai, China), while HPLC grade methanol and acetonitrile were purchased from Merck (Darmstadt, Germany). Ultrapure water was prepared using a Milli-Q system (Bedford, USA). The PAHs including fluorene (Fl), fluoranthene (FlA), pyrene (Pyr), benzo[a]anthracene (BaA), benzo[b]fluoranthene (BbF) and benzo[k]fluoranthene (BkF) were purchased from Acros Organics (NJ, USA). Stock solutions of PAH mixtures containing Fl (0.75 µg mL$^{-1}$), FlA (4 µg mL$^{-1}$), Pyr (2 µg mL$^{-1}$), BaA (0.75 µg mL$^{-1}$), BbF (0.75 µg mL$^{-1}$), and BkF (0.2 µg mL$^{-1}$) in methanol were stored at 4°C in darkness. Working standard solutions were prepared by diluting the stock solutions with ultrapure water to required concentrations for the SPME procedure.

2.2 Instrumentation

HPLC analysis of PAHs was performed with a Shimadzu (Tokyo, Japan) LC-20AT liquid chromatograph, equipped with a Shimadzu RF-10A XL fluorescence detector (FLD) and a CTO-10AS VP column oven. A Diamonsil C$_{18}$ (2) column (250 mm × 4.6 mm, particle size 5 µm; Dikma Technologies, CN) was used for separation. The mobile phase was composed of water (A) and methanol (B) with a constant flow rate of 1 mL min$^{-1}$. The elution gradient was as follows: 15 to 25 min, from 75 to 90% (B), and 25 to 32 min, from 90 to 75% (B). The excitation and emission wavelengths were programmed and are presented in Table S1.$^\dagger$

2.3 Preparation of TiO$_2$ nanotube/Ti wires

The titanium wire with TiO$_2$ nanotube arrays (TiO$_2$ NTs/Ti wire) on the surface was prepared by potentiostatic anodization of a titanium wire in a two-electrode electrochemical cell with a graphite slice as the counter electrode in a 0.5 wt% HF solution at 15 V applied by a DC power supply for 10 min at room temperature. Prior to anodization, the titanium wire was thoroughly washed with ethanol and ultrapure water in the ultrasonic bath, then dipped into the diluted HF solution for half a minute to eliminate the oxide, and finally washed with ultrapure water. After the anodization, the TiO$_2$ NTs/Ti wire was washed with ultrapure water immediately and then dried in air.

2.4 Preparation of the SPME fiber

The sol solution was prepared by mixing 80 µL of TEOS, 120 µL of C$_{18}$-TMS, 5 µL of H$_2$O, and 40 µL of TFA in a polypropylene tube. This mixture was thoroughly vortexed for 5 min, and then ready for fiber coating. One centimeter of the TiO$_2$ NTs/Ti wire was vertically dipped into the sol solution for 20 min and then placed in a desiccator overnight at room temperature. All fibers were conditioned at 120°C for 2 h prior to use. Surface structure and composition were analyzed using a scanning electron microscope (S-4800, Hitachi, Tokyo, Japan) and a Fourier transform infrared spectrophotometer (WQF-410, Braic Crop, China).

2.5 SPME procedure

The SPME fiber was assembled into a commercial device (Supelco, Bellefonte, CA) and was conditioned for 30 min in methanol before use. The extraction was performed in a 25 mL glass vial containing 20 mL sample solution. The SPME fiber was directly immersed in the sample solution for 1 h under constant stirring at room temperature to extract the analytes. After extraction, the SPME fiber was dipped in 500 µL acetonitrile to desorb analytes for off-line HPLC analysis.

2.6 River water samples

River water samples were collected from the Xiangjiang River (Changsha). The collected river water samples were filtered through a 0.45 µm cellulose membrane immediately after sampling and stored in amber glass bottles at 4°C.

3 Results and discussion

3.1 Characterization of the SPME fiber

In the sol–gel technology for the preparation of SPME fiber, an activated substrate surface with numerous silanol groups is essential for condensation reactions between silanol groups and hydroxyl-terminated sol–gel mixtures. Generally, metal substrates are oxidized to form an oxide surface for condensation reactions, e.g. a spontaneous oxide layer on the Ti wire.\textsuperscript{28} In most cases, however, the formed spontaneous oxide layer on metal surface is thin and irregular. In this work, perpendicularly orientated and well-aligned TiO$_2$ nanotubes (TiO$_2$ NTs) were successfully fabricated \textit{in situ} on the Ti wire surface by anodization, which provides rich titanol groups for condensation reactions. Fig. 1A shows the SEM image of the Ti wire surface after anodization. Uniform TiO$_2$ NTs are formed on the surface with a pore size of 70 nm and a length of 300 nm. The
was investigated by extracting hydrophobic PAHs from water samples. Fig. 2 shows the extracted amounts of PAHs (expressed in peak area) with SPME fibers fabricated with different C$_{18}$-TMS content precursor solutions. The extraction efficiency follows an increasing tendency with increasing the C$_{18}$-TMS content, indicating the key role of C$_{18}$-TMS in the extraction of PAHs. Little PAHs are extracted with coatings containing no C$_{18}$-TMS. Introduction of C$_{18}$-TMS results in an increase in the hydrophobicity of the coating, and consequently the affinity to the non-polar PAHs. The highest extraction efficiency towards PAHs with four or less aromatic rings (Flu, FlA, Pyr and BaA) is obtained with 60% C$_{18}$-TMS coating SPME fiber, while the extraction efficiency toward BbF and BkF which possess five aromatic rings increases with increasing the C$_{18}$-TMS content. These results indicate that the extraction efficiency is dependent on the interaction between the target compounds and sol–gel coating, and consequently dependent on the properties of the coating.

Furthermore, the thickness of the fiber coating also plays an important role in the extraction capability. Generally, a desirable coating thickness can be obtained by multiple dipping procedures. In this work, however, it was found difficult to increase the coating thickness by multiple dipping procedures. The reason is probably because after the first coating the surface became more hydrophobic with less surface silanol groups for sol–gel reactions, making the following coating processes difficult. A relatively high precursor concentration of sol was therefore used (refer Section 2.4) to achieve a coating thickness of ca. 2 μm which was estimated from the SEM image. Since the multiple dipping procedures failed to significantly increase the coating thickness, the extraction capability of the multi-dipped fiber was not significantly enhanced compared with the one-dipped fiber. In the following work, all the SPME fibers were prepared by the one dipping procedure.

3.2.2 Thermal Stability. The thermal stability of the SPME fiber was evaluated by subjecting it to high temperatures in the range of 240–320 °C followed by using it to extract PAHs from...
water samples. As observed in Fig. S2,† there is no loss of extraction efficiency of the fiber at the investigated temperatures, indicating a high thermal stability of the fiber. The high thermal stability suggests the presence of chemical bonding between the sol-gel coating and TiO$_2$ NTs/Ti substrate. This characteristic is very important when it is used to couple with GC analysis because it allows a high desorption temperature and consequently a short desorption time, thus minimizing or eliminating carry-over effects.

### 3.2.3 Organic solvent stability

The stability of the proposed fiber against organic solvents was evaluated by comparing its extraction efficiency before and after immersing it in organic solvents overnight. The investigated solvents include n-hexane, dichloromethane, acetone, and methanol representing polar to non-polar solvents. As shown in Fig. 3, the extracting capability toward PAHs is not reduced after overnight exposure to the investigated solvents, indicating a good stability against organic solvents. It is known that the commercial 100 μm PDMS fiber prepared by physically attaching on the substrate swells in organic solvents and is then easy to strip off from the substrate. The proposed SPME fiber shows a higher solvent stability than the commercial 100 μm PDMS fiber due to the strong adhesion on the substrate through chemical bonding.

### 3.2.4 Durability

The lifetime of the SPME fiber is very important for applications in real samples. As unbreakable Ti wire was used as the substrate, the breakage problem which usually occurs in commercial fused silica fibers was successfully solved. The lifetime of the fiber is therefore mainly dependent on its resistance to high temperatures and organic solvents. As investigated above, the chemical bonding of the coating promises the SPME fiber a high stability against high temperatures and organic solvents, and a high durability has been achieved. Fig. 4 shows the extraction capability of the fiber after subjecting to different cycles of 5 min immersion in acetonitrile and then 2 min heating at 300 °C each time before extraction of PAHs. The extracted amounts of PAHs are not decreased up to 100 cycles.

#### 3.3 Comparison of the suggested SPME fiber with commercial PDMS fibers

The extraction efficiency of the proposed SPME fiber in the extraction of PAHs was compared with the commercial PDMS fibers (7 and 100 μm) with the results presented in Fig. 5. These two commercial PDMS fibers with different coating thicknesses (7 and 100 μm) are suitable for analyzing non-polar or less-polar compounds, e.g., PAHs. As shown in Fig. 5, the 100 μm commercial PDMS fiber shows much higher extraction capability toward the analytes than the 7 μm commercial PDMS fiber, obviously due to the thicker coating. Unexpectedly, the proposed SPME fiber, even with a thinner coating of around 2 μm, provides comparable or even slightly higher extraction efficiency than the 100 μm commercial PDMS fiber toward all the investigated PAHs except for Flu.

In order to elucidate the high extraction efficiency of the proposed SPME fiber, the extraction kinetics of the proposed SPME fiber and the 100 μm commercial PDMS fiber were investigated. As shown in Fig. 6, both the SPME and PDMS
fibers show similar extraction kinetics toward all the investigated PAHs except for Flu. The extraction equilibrium of Flu is fast established on the SPME fiber due to the low extraction capacity, while the extraction equilibrium of Flu on the PDMS fiber is not reached within 1 h. That is why the extraction efficiency of SPME toward Flu is lower than the 100 μm PDMS fiber. Similar results are also observed in the extraction of BkF and BbF. These two compounds show high extraction kinetic rates and low extraction efficiencies on both SPME and PDMS fibers.

3.4 Optimization of SPME

The proposed SPME fiber coupled with HPLC-FLD was used for extracting PAHs from water samples. To achieve the best extraction efficiency, the effects of extraction time, ionic strength and desorption conditions on the SPME efficiency were studied and optimized.

SPME is a non-exhausted extraction technique and the maximum adsorbed amount of analyte is obtained when the extraction equilibrium is reached. The extraction time profile of the proposed SPME fiber for extraction of PAHs is shown in Fig. 6A. The extracted amounts (corresponding to the peak areas) of PAHs increase significantly with the increase of the extraction time within 60 min except for Flu, BbF and BkF, the extraction of which reaches equilibrium at around 20 min. In order to obtain higher analysis sensitivity, 60 min was chosen as the operational extraction time for the subsequent experiments.

Addition of salt to the sample solution can either increase or decrease the extraction efficiency via a salting-out or salting-in effect, which depends on the nature of analyte. The effect of ionic strength on the extraction efficiency of the proposed fiber for the extraction of PAHs was investigated by adding NaCl (0–30%, w/v) into the sample solution. As shown in Fig. 7, the extraction efficiency reaches the maximum at 10% NaCl and then decreases with further addition of NaCl to the sample solution. Thus, 10% NaCl was added to the sample solution in the subsequent experiments.

Desorption conditions were also studied to completely desorb analytes extracted to the fiber coating and avoid cross contamination. Desorption solvents including methanol, acetonitrile, acetone and n-hexane were investigated and the results are presented in Fig. S3.† It can be found that using acetonitrile as a desorption solvent can result in better extraction efficiency than the others. Therefore, acetonitrile was chosen as the desorption solvent. The desorption time ranging from 5 min to 30 min was also studied. As can be seen in Fig. S4,† all of the analytes are desorbed off the fiber coating at about 20 min. So, 20 min was adopted as the desorption time.

3.5 Analytical performance and application

HPLC-FLD was used for the determination of PAHs in both standard solutions and real water samples. A C₁₈ column was used to separate six structurally similar PAHs and gradient elution was performed to achieve enhanced separation selectivity and reduce analysis time. The fluorescence excitation and emission wavelengths of the six PAHs are different from each other, thus a wavelength program was adopted to obtain the maximum sensitivity for each analyte. The HPLC conditions were as follows:

- Flu: 210 nm (excitation) and 270 nm (emission);
- FIA: 270 nm (excitation) and 300 nm (emission);
- Pyr: 220 nm (excitation) and 290 nm (emission);
- BaA: 240 nm (excitation) and 270 nm (emission);
- BbF: 220 nm (excitation) and 290 nm (emission);
- BkF: 240 nm (excitation) and 270 nm (emission).

The detection limits of the six PAHs were as follows (in ng mL⁻¹): Flu, 0.38; FIA, 2.0; Pyr, 1.0; BaA, 0.38; BbF, 0.38; and BkF, 0.10.

Fig. 6 Extraction profiles of (A) the proposed SPME fiber and (B) 100 μm commercial PDMS fiber toward PAHs under the same conditions: desorption solvent, acetonitrile; desorption time, 20 min. Other conditions are the same as Fig. 2.

Fig. 7 The effect of salt concentration on the extraction efficiency of the proposed fiber. Experimental conditions: extraction time, 60 min; desorption solvent, acetonitrile; desorption time, 20 min. The desorption solvent volume (500 μL) was concentrated to 100 μL by N₂. Spiked concentration: Flu, 0.38 ng mL⁻¹; FIA, 2.0 ng mL⁻¹; Pyr, 1.0 ng mL⁻¹; BaA, 0.38 ng mL⁻¹; BbF, 0.38 ng mL⁻¹; and BkF, 0.10 ng mL⁻¹.
were optimized before the SPME procedure and the detailed mobile phase conditions are presented in Section 2.2.

The analytical features including detection limits (LODs), linear range and correlation coefficients for the extraction of PAHs with the proposed fiber are summarized in Table 1. Under the optimized conditions, linear ranges of the SPME-HPLC-FLD method are obtained by the determination of a series of mixed standard solutions. Good correlation coefficients ($R^2$ 0.9932–0.9996) for all of the analytes are achieved. The LODs, defined as three times the baseline noise, vary from 0.003 to 0.025 $\mu$g L$^{-1}$.

The repeatability of single fibers was evaluated by extracting spiked water samples under the same conditions for 5 replicates, giving a relative standard deviation (RSD) of $<7.5\%$. The fiber to fiber repeatability (using 3 different fibers from the same batch) has a RSD of $<14.2\%$.

In order to evaluate the practical applications, the proposed SPME fiber was applied in the extraction of PAHs from Xiangjiang River (Changsha, Hunan province) water samples. The collected water samples were filtered through a 0.45 $\mu$m cellulose membrane, and then spiked with the stock solution of PAHs. The unspiked water sample and spiked water sample were analyzed by the proposed SPME-HPLC-FLD method ($n=3$), and their typical chromatograms are presented in Fig. 8. As can be seen, Flu is detected in Xiangjiang River water samples with a concentration of 0.053 $\mu$g L$^{-1}$, while the others are under the LODs. The recoveries were in the range of 85.3–101.8%. These satisfactory recoveries indicate no significant matrix effects for the environmental water samples.

4 Conclusion

A novel, unbreakable SPME fiber was fabricated by sol–gel technology using TEOS and C$_{18}$-TMS as co-precursors and TiO$_2$ NTs/Ti wire as the substrate. The sol–gel coating exhibited hydrophobic characteristics, favoring the extraction of non-polar compounds. Though the coating thickness is only ca. 2 $\mu$m, the extraction efficiency toward PAHs is compared favourably to the 100 $\mu$m commercial PDMS fiber. The proposed fiber exhibited high thermal and organic solvent stability, as well as satisfactory durability, extraction repeatability and preparation reproducibility.

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References


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<th>Reproducibility ($n=3$, %) (fiber to fiber)</th>
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* Experimental conditions: extraction time, 60 min; salt addition, 10% (w/v); desorption solvent, acetonitrile; desorption time, 20 min; desorption solvent volume (500 $\mu$L) was concentrated to 100 $\mu$L by $N_2$. * Correlation coefficient. * LODs were estimated as the concentrations where $s/n = 3$.

Fig. 8 HPLC chromatograms of unspiked (a) and spiked (b) river water samples extracted by the SPME fiber for PAHs: (1) Flu; (2) FIA; (3) Pyr; (4) BaA; (5) BbF; and (6) BkF. Spiked concentration: Flu, 0.38 ng mL$^{-1}$; FIA, 2.0 ng mL$^{-1}$; Pyr, 1.0 ng mL$^{-1}$; BaA, 0.38 ng mL$^{-1}$; BbF, 0.38 ng mL$^{-1}$; and BkF, 0.10 ng mL$^{-1}$. The peak of x was not identified.