Talanta 185 (2018) 23–29

Contents lists available at ScienceDirect

Talanta

journal homepage: www.elsevier.com/locate/talanta

Solid-phase microextraction of volatile organic compounds in headspace of PM-induced MRC-5 cell lines

JingHong Li[a], Nan Chen[b], YiPing Tian[b], Hui Xu[a,*]

[a] Key Laboratory of Pesticide & Chemical Biology, Ministry of Education, Institute of Analytical Chemistry, College of Chemistry, Central China Normal University, Wuhan 430079, China

[b] Hubei Provincial Environmental Monitoring Center Station, Wuhan 430072, China

ARTICLE INFO

Keywords:
Solid phase microextraction-gas chromatography-mass spectrometry
Volatile organic compounds
MRC-5
Particulate matter
Graphene oxide

ABSTRACT

The exploration of volatile organic compounds (VOCs) produced by cell lines may be a powerful and non-invasive tool for the study of the health risk of human exposure to atmospheric particulate matter (PM). In this work, we developed a sensitive solid phase microextraction-gas chromatography-mass spectrometry method (SPME-GC-MS) to analyze VOCs in breathed gas of PM2.5-induced human embryonic fibroblast cell line (MRC-5). A novel graphene oxide/polyaniline/polydopamine (GO/PANI/PDA) coating was prepared on a stainless steel wire via electrochemical deposition and self-polymerization for the first time. The GO/PANI/PDA coating exhibited high extraction efficiency, good thermal stability (> 380 °C), excellent mechanical stability as well as long service time (> 150 times). Parameters that may affect the results were optimized systematically. Under the optimal conditions, VOCs including benzene series, aldehydes and alkane were detected with low limit of detection (0.2–2.0 μg L^{-1}) and good correlation (correlation coefficients above 0.9922). The relative standard deviations of within-day and between-day were 1.1–8.4% and 0.2–11.2%, respectively. Satisfactory recoveries of 82–117% indicated good repeatability of the method. The method has been successfully applied for the determination of target VOCs in the headspace gas of PM2.5-induced MRC-5 cell. And it is expected to provide an alternative tool for the study of cytotoxicology of atmospheric particulates.

1. Introduction

Atmospheric particles matter (PM) especially PM2.5 that is with aerodynamic diameters of 2.5 μm and less in size have a serious risk on human health [1–3], because it is easy to reach distal regions of lung and exert bad effect [4–6]. It was reported that the exposure to air particulates could promote the exacerbation of allergic and respiratory diseases [7]. PM-induced diseases are often related with prooxidation and antioxidation imbalance [8], and the excess reactive oxygen species can bring to oxidative stress, which further leads to protein and DNA peroxidation, lipid peroxidation as well as the content change of volatile organic compounds (VOCs) in exhaled breath [9]. So, the study of VOCs changes in PM-induced lung cell lines might be a new strategy to investigate cytotoxicology of PM 2.5 on human bodies.

The exhaled gas from cells or bodies contains hundreds of compounds [9], so suitable sample preparation techniques are needed for the selective enrichment of trace amount of VOCs markers in breathed gas [10,11]. In order to eliminate complex matrices, enrich the targets and obtain good analytical data, a new sample preparation method, solid-phase microextraction (SPME) was proposed by Pawliszyn et al. in the early 1990s [12]. SPME is based on the absorption-desorption distribution equilibrium of analytes between matrix and coatings on fibers. It only needs minimized organic solvents in extraction and could combine sampling, preconcentration and introduction in one step, thus achieves simplification of procedure [12,13]. Nowadays, SPME has become one of the most widely used pretreatment methods for the extraction of volatile and semi-volatile substance [12–17]. The SPME sorbents are of great importance for extraction, because the recommended temperature, the ability of solvent resistance, the cycle times as well as mechanical stability of coatings can influence sensitivity and accuracy of a method [16,17]. Commercial SPME fibers are available for common applications, but the limited kinds of coatings cannot meet the requirements of analysis [13]. Many researches contribute to the discovery of novel coating materials.

Polyaniline (PANI), a kind of conducting polymer that can be electrochemically synthesized by monomers and designed with various oxidation forms, has been extensively applied as sorbents [18]. The chain-like PANI can exhibit strong interaction to aromatic compounds due to the existence of large π–π bonds [19,20]. But PANI was heat labile when it was used for thermal desorption in GC port [20]. Self-

* Corresponding author.
E-mail address: huixu@mail.ccnu.edu.cn (H. Xu).

https://doi.org/10.1016/j.talanta.2018.03.041
Received 3 October 2017; Received in revised form 7 March 2018; Accepted 14 March 2018
Available online 15 March 2018
0039-9140/

doped PANI or doping with dopants can be an efficient method to solve the problem. Carbon materials as dopants have aroused great interest during the few years, because they can be used to improve the thermal stability of PANI [21,22]. Graphene (GO) is an emerging carbon material that possesses the thickness of one carbon atom arranged in aromatic macromolecule sheet. All the features of GO including good chemical and mechanical stability, unique thermal stability, and especially high surface-to-weight ratio make it an attractive candidate for adsorption [23]. For example, Mehdinia et al. prepared a graphene-polyaniline fiber which showed high selectivity to hydrophobic compounds [24]. However, the GO/PANI coating may be unsuitable for the enrichment of hydrophilic compounds, due to its strong hydrophobicity.

Polydopamine (PDA) is a novel hydrophilic material inspired by bio-adhesive ability. The self-polymerization of dopamine can take place in mild alkaline solutions, and the resultant PDA which contains rich catechol and amino groups could present strong adhesion on variety of substrates and act as highly stable film [25,26]. Besides, it is environmentally stable and biocompatible [27]. Many researches have revealed that PDA could enhance the extraction efficiency when it was used as additive for SPME coatings [28–30].

In this work, we developed a sensitive and eco-friendly SPME-GC-MS method in which graphene oxide/polyaniline/polydopamine was applied as a novel fiber coating. The surface characterization, thermal stability, lifetime and extraction ability of composite coating were investigated. Several parameters that affected extraction and desorption efficiency were optimized. Nine VOCs including benzene, ethylbenzene, nonanal, chlorobenzene, benzaldehyde, undecane, toluene, pyridine and phenylacetaldehyde were proposed to be potential volatile biomarkers of lung cancer in previous reports [13,31–36]. And they were selected as model analytes to investigate the effect of PM2.5 on MRC-5 cell lines. The developed method was applied to the determination of VOCs in headspace gas of PM2.5-induced MRC-5 cell lines.

2. Materials and methods

2.1. Chemicals and materials

Chlorobenzene (99.5%), benzaldehyde (99%) and benzeneacetaldehyde (97.5%) were gained from AccuStandard Inc. (USA). Benzene (99.5%), toluene (98%) and undecane (99%) were obtained from TCI Co., Ltd. (Shanghai, China). Nonanal (97%) and ethylbenzene (99.7%) were purchased from Alfa Aesar (Tianjin, China) and Amethyst Chemicals (Beijing, China), respectively. HPLC grade ethanol from Biologic-Reagents (USA) and ultra-pure water from Sartorius Stedim Biotech (ariums® pro Ultrapure Water Systems, Gottingen, Germany) were used throughout the experiment. Aniline, graphene oxide (GO) and dopamine hydrochloride (98%) were obtained from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China), Xfnano, Inc. (Nanjing, China), and Aladdin Industrial Corporation (Shanghai, China), respectively. The stainless steel wire (0.25 mm diameter, 23 cm length) was 304-stainless steel (Hengyang Metal Co., Ltd., Shenzhen, China). Minimal essential medium (MEM) and Trypsin (Life, USA), phosphate buffered saline (PBS, Sigma-Aldrich, Germany), fetal bovine serum (FBS, Gibco, USA), streptomycin and penicillin (Hyclone, USA) were used for cell cultures.

2.2. Preparation of standard solution

Standard stock solution of nine compounds (including benzene, pyridine, chlorobenzene, ethylbenzene, toluene, benzaldehyde, benzeneacetaldehyde, nonanal and undecane) was prepared in ethanol at the concentration of 1650 μg mL^{-1}. The daily working solutions were obtained by diluting the stock solution in 10 mL volumetric flasks with ethanol as solvent. All of the solutions were stored at − 20 °C. The standard gas involving the mentioned VOCs was prepared via evaporation of 3 μL working solution in a 25 mL gastight vial (Agilent, USA).

2.3. Preparation of graphene oxide/polyaniline/polydopamine fiber

The graphene oxide/polyaniline/polydopamine (GO/PANI/PDA) fiber was prepared in the following steps. (1) 30 mg GO was uniformly dispersed in 10 mL pure water with the continuous ultrasonication for three hours to form a homogeneous suspension. Then, aniline were added to the above suspension to obtain the electrolyte dispersions which contained 3 mg mL^{-1} GO and 0.1 mol L^{-1} aniline. The mixed dispersions was deoxygenated with N_2 for 5 min (2) Prior to preparation of coating, two stainless steel wires (outside diameter: 0.2 mm) were cleaned with acetone, ethanol and pure water in ultrasonic device. Graphene oxide/polyaniline (GO/PANI) coating was in-situ deposited on the outer surface of the clean stainless steel wire via electrochemical deposition in electrolyte solution. The deposition experiment was carried out with a simple homemade electrochemical system, which consisted of a 9 V Panasonic battery (direct-current power), two stainless steel wires (acted as anode and cathode), and two conductors. After 8 s of deposition under 9 V direct current, black composites formed on the outer surface of stainless steel anode with the length of 1.5 cm, the fiber was took out and washed with methanol and pure water to eliminate impurities. (3) The procedure was repeated for five times to provide a desirable thickness and the fiber was dried at 60 °C before the modification of polydopamine. (4) 160 mg dopamine hydrochloride was dissolved in 80 mL Tris buffer solution (pH = 8.5, 10 mM) and the obtained fiber coating was immersed in the solution under continuous stirring for 24 h. After self-polymerization of dopamine, the prepared fiber was washed with methanol and distilled water to remove the unreacted dopamine monomer, then it was conditioned at 250 °C under N_2 for 1 h. Finally, the prepared fiber was installed in a 5 μL microsyringe as a SPME holder [15].

2.4. Instrumentation

The GC-MS-QP 2010 plus (Shimadzu, Japan) system was used for VOCs analysis. Helium was employed as carrier gas and its flow rate was set at 1.68 mL min^{-1}. The temperature of ion source and interface were 200 °C and 240 °C, respectively. The energy of electron impact ionization (EI) was 70 eV. The separation of VOCs was performed on a CP-Pora bond Q column (25 m × 0.32 mm × 5 μm, Varian Chrompack, USA). The column temperature was firstly held at 100 °C for 0.5 min, then increased to 280 °C at the rate of 8 °C min^{-1}, and maintained for 4 min. The quantitative data was obtained by selected ion monitoring (SIM) mode. Commercially available SPME fibers (Polyacrylate (PA), Polydimethylsiloxane (PDMS)) were purchased from Supelco (USA). The Retention time, ions to identify and quantify VOCs are shown in Table S1 (see in Supporting information).

The morphology of the prepared fiber coating was observed by scanning electron microscope (JSM-IT300, Japan). Fourier Transform infrared spectra were recorded on a Spectrum two Spectrometer (PerkinElmer, USA).

2.5. Preparation of real samples

2.5.1. Cell cultures

MRC-5 obtained from Chinese Academy of Sciences Cell Bank (Shanghai, China) were cultured in MEM medium with 10% FBS, 3% penicillin (10,000 U mL^{-1}) and 3% streptomycin (10,000 μg mL^{-1}) in a 37 °C humidified atmosphere containing 5% CO_2.

2.5.2. PM2.5 treatment and cell viability measurement

PM2.5 was collected from 13 mm-diameter sample filtration membranes of TEOM 1405D Ambient Particulate Monitor (Thermo, USA). After two weeks' collection at Wuhan (Hubei, China), every filter was