Boron nitride nanotubes matrix for signal enhancement in infrared laser desorption postionization mass spectrometry

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ABSTRACT

The nanomaterials function as the substrate to trap analytes, absorb energy from the laser irradiation and transfer energy to the analytes to facilitate the laser desorption process. In this work, the signal intensity and reproducibility of analytes with nanomaterials as matrices were explored by laser desorption postionization mass spectrometry (LDPI-MS). Herein, the desorbed neutral species were further ionized by vacuum ultraviolet (VUV, 118 nm) and analyzed by mass spectrometer. Compared with other nanomaterial matrices such as graphene and carbon nanotubes (CNTs), boron nitride nanotubes (BNNTs) exhibited much higher desorption efficiency under infrared (IR) light and produced no background signal in the whole mass range by LDPI-MS. Additionally, this method was successfully and firstly exploited to in situ detection and imaging for drugs of low concentration in intact tissues, which proved the utility, facility and convenience of this method applied in drug discovery and biomedical research.

1. Introduction

Nanomaterials, such as graphene, carbon nanotubes (CNTs) and boron nitride nanotubes (BNNTs), possess large surface area, excellent optical absorption and super electrical properties [1–3]. With the inherent superiorities, they were considered as prominent materials in energy absorption, storage and transfer due to their unique π-conjugated structures. Because of these characteristics, nowadays nanomaterials have played significant effect on the development of novel mass spectrometer [4]. In recent years, considerable efforts have been devoted to eliminate matrix ion interference come from the conventional organic matrices by using different nanomaterial substances, such as graphene, carbon nanotubes (CNTs), graphitic carbon nitride (g-C_3N_4) nanosheet and hexagonal boron nitride (h-BN) nanosheets [5–8]. The nano matrices were not only applied in vacuum ionization method but also used in atmospheric pressure ionization strategy such

as ambient plasma assisted laser desorption ionization mass spectrometry (PALDI-MS) [9]. One of the reasons why the nano matrices can increase the mass signal intensity is that they function as the substrate to trap analytes efficiently [10]. Furthermore, they can improve the reproducibility of intra- and inter-sample spots for their ability of uniform crystallization. However, the main purpose of above experiments was to eliminate the background signal and little efforts were payed to study the signal enhancement mechanism.

According to previous research, boron nitride (BN) based nanomaterials have high capacity of adsorption and no background signal [7]. Similar to graphite, diamond and CNT phases, BN has tubular, layered and cubic structure [11]. Boron nitride nanotube (BNNT) is structurally analogous to CNT with carbon atoms replaced by boron and nitrogen atoms. To date, several modern measuring techniques have been used to verify its incomparable characteristics. The thermal conductivity of BNNTs was measured by using a pulsed photothermal technique to be

about 100 W m^{-1} K^{-1}, making them one of the best electrically insulating thermal conductors [12]. Atomic force microscopy (AFM) and high resolution transmission electron microscopy (HRTEM) measurements also revealed that the structures of individual BNNTs can survive at up to 850 °C in air and capture the signs of their structural degradation at 900 °C or above [13].

Though it is hard to synthesize this nanomaterial and the price of it is a little high, possessing some excellent physical and chemical properties make it being widely utilized nowadays. It has been proposed to protect metals against oxidation and corrosion at high temperatures [14]. BNNTs and their functionalization via quinuclidine-3-thiol with gold nanoparticles also have been used for enhancing the efficiency of the monolithic columns in high performance liquid chromatography (HPLC) [15]. However, any efforts to evaluate BNNTs as a novel matrix used in mass spectrometry (MS) have not been well-investigated so far, especially used in laser desorption postionization mass spectrometry (LDPI-MS) for the enhancement of mass signals.

LDPI-MS, as a state-of-the-art approach, has played great significance in studying cosmogeochemistry [16], exploring the non-volatile organics in mineral from other planetary bodies [17,18], analyzing of drugs, rare earth element residues [19], organic acid metabolites and imaging of biological material [20]. In LDPI, the desorption and ionization of analytes are separated spatio-temporally: the first laser desorbs surface material and the second 118 nm beam ionizes the desorbed neutral gaseous plume [21,22]. This is unlike the surface assisted laser desorption/ionization (SALDI) where the substrate serves as a source of thermal energy to desorb the analytes [23]. The two distinct steps would promote the overall sensitivity and provide more easily interpretable results [24]. Moreover, this selective ionization would simplify mass spectra, minimize the interference of the background signal, increase the linearity and improve the ratio of signal-to-noise [25]. What's more, LDPI-MS has the advantages in the detection of low mass range molecules [26]. In this study, we investigated the signal intensity and reproducibility influenced by nanomaterials as matrix added on samples in LDPI-MS, as well the desorption efficiency of desorption laser at different wavelengths which have never been done before. BNNTs, as a novel matrix, were sought out and found to exhibit higher desorption efficiency under infrared laser and background-free in the whole mass range by LDPI-MS. Finally, this method was used to localize the drugs at low concentration in the kidney tissue, which verified the potential applications of this method for real samples.

2. Materials and methods

2.1. LDPI-MSI apparatus

The schematic diagram of the home-built apparatus was elaborated in Fig. 1. The principle components consisted of a custom-designed linear time of flight mass spectrometer (TOF-MS) with the vacuum chamber pressure was about 10^{-7} Torr and a third harmonic generation (THG) cell. A ternary 36 mm diameter position-sensitive detector (microchannel plate, MCP) was used to collect the ion signals. Each saved mass spectrum was the average of 532 laser shots acquired by a digital oscilloscope (DPO4102B-L, Tektronix Inc., Beaverton, OR, USA). A DG535 digital delay gate generator (Stanford Research Systems, Sunnyvale, CA, USA) was employed to set the relative delay between the desorption and postionization laser pulses. A translatable sample plate for mass spectrometry imaging (MSI) was realized by an X-Y-Z stage with three motors.

In this experiment, a 532/1064 nm Nd:YAG laser (Minilite, Continuum Inc., San Jose, CA, USA) with a 10 Hz operational repetition rate was focused to ca. 150 μm onto the sample surface by a plano-convex lens (f = 50 mm) to desorb the analytes. The optimized desorption laser energy shot at the irradiated area was approximately 300 μJ. Tunable infrared (IR) radiation in the 1500–5000 nm
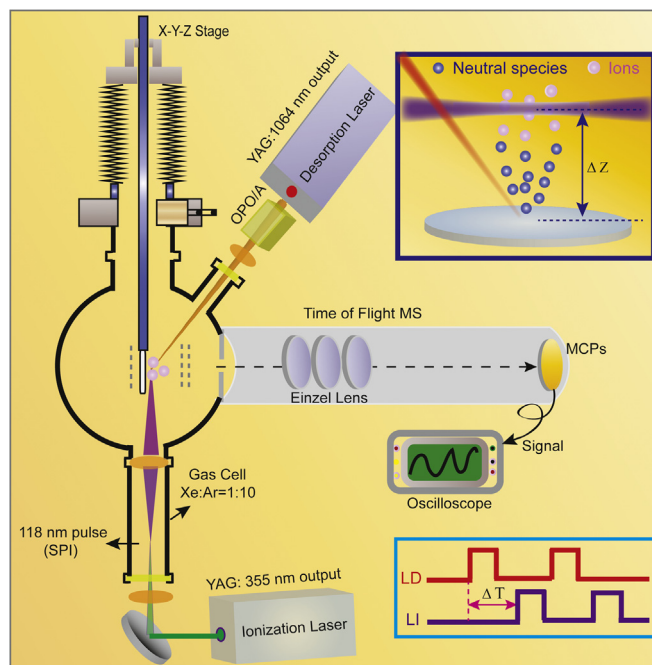

Fig. 1. Schematic drawing of the laser desorption postionization mass spectrometer imaging apparatus and the sample hold with an X-Y-Z stage. ΔZ represents the distance between the sample surface and VUV beam and ΔT represents the delay time between the desorption and ionization lasers.

wavelength range was generated from an optical parametric oscillator/optical parametric amplifier (OPO/OPA) system (Laser Vision, Bellevue , WA, USA) which was pumped by a 1064 nm Nd:YAG laser (Surelite EX, Continuum Inc., San Jose, CA, USA). Vacuum ultraviolet (VUV 118 nm, 10.5 eV) photons were generated by frequency tripling another Nd:YAG 355 nm laser (INDI-40-10YAG, Spectra-Physics Inc., Santa Clara, CA, USA) in a THG cell filled with high-purity nonlinear medium of a ~ 200 Torr in phase-matched 1:10 Xe/Ar gas. Briefly, the 10 Hz, 355 nm laser (ca. 30 mJ/pulse) was focused with a quartz lens (f = 25 cm) into THG cell to generate 118 nm photons [27]. The separation of the VUV photons and 355 nm beam was caused by the large difference between the refractive indices of the MgF$_2$ lens at the YAG third harmonic (n = 1.38 at 355 nm) and ninth harmonic (n = 1.67 at 118 nm) [28]. On the basis of approximate conversion efficiency of 10^{-5} reported in the literature, the VUV pulse energy was thought to be about 300 nJ though it has not been measured [28,29]. The VUV beam paralleled to the corundum sample plate was brought into the vacuum chamber where it was focused to a ca. 0.1 mm^2 focus point above the desorption region. Meanwhile, the sample plate run horizontally to ensure the topography changed slightly while scanning the tissue sections [24].

2.2. Reagents

Graphene, CNTs and BNNTs were purchased from Beijing DK nano S &T Ltd (Beijing, China). Levamisole (C$_{11}$H$_{12}$N$_2$S), 1,5-naphthalenediamine (C$_{10}$H$_{10}$N$_2$), 1,5-diaminoanthrachinon (C$_{14}$H$_{10}$N$_2$O$_2$), pyrimethamine (C$_{12}$H$_{13}$ClN$_4$) and 2,5-dihydroxybenzoic acid (DHB, C$_7$H$_6$O$_4$) were obtained from Sigma-Aldrich (St. Louis, MO, USA). Alcohol (C$_2$H$_5$OH), sodium hydroxide (NaOH), sodium chloride (NaCl), dipotassium phosphate (K$_2$HPO$_4$) and sodium hydrogen phosphate (Na$_2$HPO$_4$) were obtained from Guangzhou Chemical Reagent Factory (Guangzhou, China). The buffer solution used in the experiment was the phosphate-buffered saline (PBS , pH 7.4) which contained 100 mM NaCl and 10 mM Na$_2$HPO$_4$/K$_2$HPO$_4$. The embedding material (Optimal Cutting Temperature, OCT) was obtained from Yi Sheng Biotechnology