Science of the Total Environment 554–555 (2016) 246–252

Contents lists available at ScienceDirect

Science of the Total Environment

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

Surface-enhanced Raman scattering detection of silver nanoparticles in environmental and biological samples

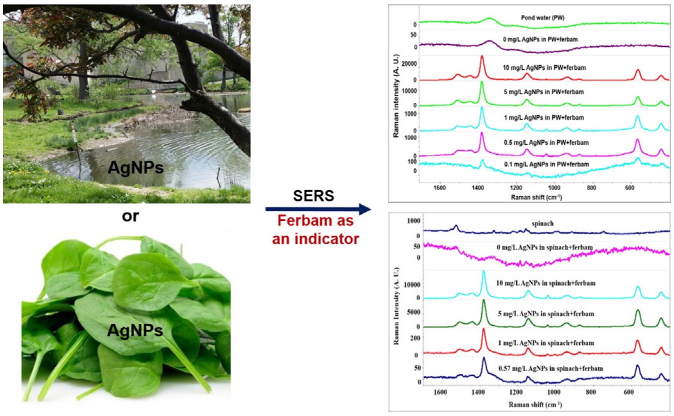
Huiyuan Guo [a], Baoshan Xing [a,*], Leigh C. Hamlet [a], Andrea Chica [a], Lili He [b,*]

[a] Stockbridge School of Agriculture, University of Massachusetts, Amherst, MA 01003, USA
[b] Department of Food Science, University of Massachusetts, Amherst, MA 01003, USA

HIGHLIGHTS

- Ferbam was found to be the best indicator for SERS detection of AgNPs.
- SERS was able to detect AgNPs in both environmental and biological samples.
- Major components in the two matrices had limited effect on AgNP detection.

GRAPHICAL ABSTRACT



SERS signal intensity of ferbam indicates the concentration of AgNPs.

ARTICLE INFO

Article history:
Received 1 January 2016
Received in revised form 12 February 2016
Accepted 12 February 2016
Available online xxxx

Editor: D. Barcelo

Keywords:
SERS
AgNPs
Environmental water
Biological samples
Matrix components

ABSTRACT

Growing concerns over the potential release and threat of silver nanoparticles (AgNPs) to environmental and biological systems urge researchers to investigate their fate and behavior. However, current analytical techniques cannot meet the requirements for rapidly, sensitively and reliably probing AgNPs in complex matrices. Surface-enhanced Raman spectroscopy (SERS) has shown great capability for rapid detection of AgNPs based on an indicator molecule that can bind on the AgNP surface. The objective of this study was to exploit SERS to detect AgNPs in environmental and biological samples through optimizing the Raman indicator for SERS. Seven indicator molecules were selected and determined to obtain their SERS signals at optimal concentrations. Among them, 1,2-di(4-pyridyl)ethylene (BPE), crystal violet and ferric dimethyl-dithiocarbamate (ferbam) produced the highest SERS intensities. Further experiments on binding competition between each two of the three candidates showed that ferbam had the highest AgNPs-binding ability. The underlying mechanism lies in the strong binding affinity of ferbam with AgNPs via multiple sulfur atoms. We further validated ferbam to be an effective indicator for SERS detection of as low as 0.1 mg/L AgNPs in genuine surface water and 0.57 mg/L in spinach juice. Moreover, limited interference on SERS detection of AgNPs was found from environmentally relevant inorganic ions, organic matter, inorganic particles, as well as biologically relevant components, demonstrating the ferbam-assisted SERS is an effective and sensitive method to detect AgNPs in complex environmental and biological samples.

---

* Corresponding authors.
E-mail addresses: bx@umass.edu (B. Xing), lilihe@foodsci.umass.edu (L. He).

http://dx.doi.org/10.1016/j.scitotenv.2016.02.084
0048-9697/

1. Introduction

Silver nanoparticles (AgNPs) have raised concerns over their potential threat to human health and the environment due to their increasing applications and potential toxicity (Bartłomiejczyk et al., 2013; Kim and Ryu, 2013; Schluesener and Schluesener, 2013; Wijnhoven et al., 2009). Among nanotechnology-based consumer products on the market, the number of products containing AgNPs are the largest (www.nanotechproject.org). Currently in the US, there are more than 110 officially registered silver containing pesticides used for medical, agricultural, environmental and home purposes (Bergeson, 2010). Many of the registered pesticides contain nanosilver. The widespread use of AgNPs is mainly due to their higher antimicrobial activity relative to the bulk silver particles. However, the large scale applications of AgNPs increase the likelihood of human exposure and highlight the importance of thoroughly understanding AgNP fate and effects in biological systems. For example, if AgNPs are taken up by plants, further bioaccumulation into higher trophic level organisms may be initiated, which can affect the entire food chain and pose risks to humans and other organisms (Gardea-Torresdey et al., 2014; Hawthorne et al., 2014). So far, AgNPs-induced risks, such as oxidative stress, genotoxicity and apoptosis in human cells have been reported by a number of studies (Bartłomiejczyk et al., 2013; Kim and Ryu, 2013). For example, AgNPs (5–10 nm) ranging from 0.05 mg/L to 0.2 mg/L could lead to oxidative stress, DNA damage, cell cycle arrest and apoptosis in human jurkat cells after 24 h treatment (Eom and Choi, 2010). Therefore, it is important to assess the levels of AgNPs in food, agricultural and environmental samples.

However, most current techniques for characterizing and detecting AgNPs have various problems (Majedi and Lee, 2016; Tiede et al., 2008). Electron microscopy techniques, such as scanning electron microscopy (SEM) and transmission electron microscopy (TEM), have been used to image the size and morphology of AgNPs. However, these techniques require complicated sample preparation procedures, especially for biological samples. Also, they cannot identify silver species unless they are coupled with energy dispersive X-ray spectroscopy (EDS). In other words, SEM and TEM results cannot be reliably used for interpreting and predicting the environmental behavior and toxicity of AgNPs, which are chemically reactive and can undergo various transformations in the environment (Levard et al., 2012). Dispersive light scattering (DLS) and UV–vis spectroscopy can characterize AgNPs but the results may be interfered by sample matrices. Several elemental quantification techniques, including graphite furnace atomic absorption spectrometry (GFAA), inductively coupled plasma optical emission spectrometry (ICP-OES) or inductively coupled plasma mass spectrometry (ICP-MS), are capable to measure total Ag concentration, but they fail to provide the speciation information of Ag. Currently, more advanced techniques, such as single particle inductively coupled plasma mass spectrometry (sp-ICP-MS) and asymmetric flow field flow fractionation ICP-MS (AF4-ICP-MS), have been developed to straightforwardly monitor different forms of AgNPs in simple water samples. However, their sensitivity may be mitigated in terms of low concentrations of AgNPs in complex samples (Mwilu et al., 2014). Overall, three main bottlenecks are faced by current techniques: complex sample preparation, difficulty in differentiating AgNPs from other Ag species and matrix interferences. Therefore, reliable analytical methods that can meet these challenges are urgently needed.

SERS is an advanced technique that incorporates nanoscale noble metal substrates into normal Raman spectroscopy. Weak Raman signals can be improved by many orders of magnitude because the excitation of localized surface plasmon resonance (LSPR) on nanoscale-roughened surfaces can generate a large electromagnetic field which increases the Raman cross section from the molecules adsorbed to noble metal nanostructures (Haynes et al., 2005). While most researchers utilized nano silver as the substrate of SERS to analyze chemical or biological analytes, we applied SERS in an opposite way, i.e., use a compound that can bind onto the AgNPs and produce characteristic signals to indicate the presence of AgNPs. Since SERS intensity is dependent on the inherent properties of the ligand molecules as well as the interaction between AgNPs and ligands, the indicator selection is the key to develop a sensitive and effective method for analysing AgNPs in complex matrices. A good indicator should 1) produce distinct signals with high intensity at low concentrations upon mixing with AgNPs, and 2) have very strong affinity to the AgNP surface so that it can outcompete with other molecules in the sample for adsorbing on the NP surfaces or replace any ligand already on the surface of AgNPs. To date, the ability of SERS method to quantify AgNPs has not been investigated in complex sample matrices, such as environmental water and biological samples. Whether SERS method could accurately identify AgNPs in complex matrices is dependent on if the indicator could locate and adsorb onto AgNPs in the presence of potentially interfering matrix components.

Here in this study, we aimed to experimentally select and validate the best SERS indicator and further applied the indicator-assisted SERS method to detect AgNPs in real environmental and biological samples (i.e., surface water and spinach leaves). Furthermore, a detailed analysis of the matrix effects on AgNP detection by SERS was performed. As far as we know, this is the first time that the abilities of different indicator molecules for detecting AgNPs were evaluated and the indicator-assisted SERS was applied to quantify AgNPs in both environmental and biological samples, with a thorough investigation of potential interfering effects of matrix components.

2. Materials and methods

2.1. Materials

Acetone (≥99.5%), $AgNO_3$ (99.8%), ferbam, melamine (99%), sodium citrate, sodium bicarbonate, sodium sulfate anhydrous, sodium chloride, titanium oxide (98.0–100.5%) and Au slides (for SERS) were all obtained from Fisher Scientific (Pittsburgh, PA). Sodium carbonate anhydrous was ordered from J.T. Baker (Center Valley, PA). Crystal violet, rhodamine 6G (R6G, 99%), 1,2-di(4-pyridyl)ethylene (BPE, 18.223 mg/L) and humic acid (sodium salt) were purchased from Sigma-Aldrich (St. Louis, MO) while selenium dimethyldithiocarbamate was purchased from Gelest (Morrisville, PA) and ruthenium red (99.9%) was obtained from Pfaltz & Bauer (Waterbury, CT).

Pond water was collected from University of Massachusetts Amherst in October 2014. A total carbon analyzer (Shimadzu, TOC-L) was used to measure inorganic carbon content and total organic carbon content (TOC) in the pond water sample. A conductivity/pH meter (Fisher Scientific, XL200) was used to determine electrical conductivity (EC) and pH of the water sample. To analyze the total solid concentration, 50 mL of water sample was placed into a clean dish and dried in an oven at 100 °C for 24 h. The mass difference between the clean dish and the dried dish with residue was calculated to represent the total solid concentration. Fresh spinach leaves were purchased from a local supermarket (Stop & Shop, Hadley, MA).

2.2. Methods

2.2.1. Assessment of the SERS intensity of indicator molecules

To our knowledge, various dye molecules with a conjugated double bond system are very Raman active (producing high intensity peaks at low concentrations). They have been widely used in various sensors or probes for cellular and molecular diagnostics by incorporating them on or in the NPs (Huh et al., 2009). Also, molecules with multiple sulfur or nitrogen atoms can bind Au and Ag strongly and are widely used for surface modifications of Au or Ag substrate to improve their functionalities (Doering and Nie, 2003; Graham et al., 2001). Accordingly, we chose seven potentially good candidates which are dyes and/or contain multiple pyridyl, sulfur or nitrogen atoms: ferbam, crystal violet, rhodamine 6G (R6G), melamine, 1,2-di(4-pyridyl)ethylene (BPE), selenium