Sensors and Actuators B 238 (2017) 641–650

Contents lists available at ScienceDirect

Sensors and Actuators B: Chemical

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

Instantaneous detection of melamine by interference biosynthesis of silver nanoparticles

S.C.G. Kiruba Daniel, Lourdes Albina Nirupa Julius, Sai Siva Gorthi*

Department of Instrumentation and Applied Physics, Indian Institute of Science, Bangalore, India

ARTICLE INFO

Article history:
Received 5 April 2016
Received in revised form 27 June 2016
Accepted 21 July 2016
Available online 25 July 2016

Keywords:
Melamine
Interference biosynthesis
Silver nanoparticles
Instant detection

ABSTRACT

Instantaneous detection of melamine, a potential milk adulterant has been demonstrated at room temperature by means of interference biosynthesis of silver nanoparticles. The sensing mechanism is based on the colorimetric change observed during the synthesis of silver nanoparticles due to the presence of melamine added during the biosynthesis. Presence and absence of melamine led to either inhibition of nanoparticle formation or enable partial synthesis of nanoparticles which is detected spectrally. A limit of detection (LOD) of 0.1 ppm in water and 0.5 ppm in raw milk was detected by the proposed technique at room temperature. UV–vis spectroscopy and High Resolution Transmission Electron Microscopy (HR-TEM) have been used to detect the spectral Surface Plasmon Resonance (SPR) and morphological changes of synthesized silver nanoparticle with and without the presence of the analyte melamine. Further, interference synthesis based sensing of melamine was done with caffeic acid as a reducing agent which confirms the role of caffeic acid a major constituent of Parthenium leaf extract for interference biosynthesis based sensing. Melamine is detected from raw milk by interference biosynthesis based sensing after a facile milk pre-processing step. Thus the method can be converted into a workable handheld prototype for detection of melamine for in-situ field applications.

1. Introduction

Adulteration of food is one of the major problems in the food industry and their detection has become the need of the hour [1]. It will be a boon to detect food adulteration at a rapid rate within a few minutes using a simple technology to prevent health hazards. Food materials/drinks when adulterated will lead to serious health issues based on the adulterant being used [1]. Melamine is one such adulterant milk which when added to milk makes it look protein rich due to the high nitrogen content in melamine. Adulteration of milk by melamine will lead to formation of kidney stones and other renal problems in infants as well as adults [2]. Widespread melamine adulteration was reported in China in 2008 where about 54,000 children were hospitalized with few reported deaths [3]. Currently melamine is being detected commercially by means of Gas Chromatography–Mass Spectrometry (GC–MS), High Performance Liquid Chromatography (HPLC), Liquid Chromatography–Mass Spectrometry (LC–MS) and other sophisticated techniques [4]. The above mentioned centralized laboratory testing techniques require skilled personnel for conducting the test and to interpret the results obtained. Hence there is a need for the detection of melamine in a rapid, simple and affordable way.

With the advent of nanotechnology, different research groups have demonstrated the usage of silver and gold nanoparticles for sensing of melamine [5]. Most of the literature employed previously synthesized metal nanoparticles for sensing applications. Noble metal nanoparticles owing to their Localized Surface Plasmon Resonance (LSPR) have been utilized for different sensing applications [6–14]. Similarly LSPR of metal nanoparticles – Gold and Silver have been put into use for the detection of melamine. In most of the publications where melamine is detected by metal nanoparticles, two steps are followed viz – synthesis of nanoparticles followed by sensing of analytes [5]. Synthesis followed by sensing requires some time before the sensing takes place. In the current strategy sensing of the analyte by the nanoparticles will be due to the interference in the biosynthesis of nanoparticles by the analyte at room temperature. Recently biosynthesized silver nanoparticles have been used to detect melamine where, previously synthesized nanoparticles were utilized to sense melamine [15]. Interruption in synthesis of nanoparticles (gold and silver) based sensing has been previously reported for the detection of melamine using reducing agents like hydrogen peroxide, dopamine, ellagic acid [16–18] while there are no reports of a leaf extract based synthesis interruption leading to

* Corresponding author.
E-mail addresses: saisiva.gorthi@iap.iisc.ernet.in, saisiva.gorthi@gmail.com (S.S. Gorthi).

http://dx.doi.org/10.1016/j.snb.2016.07.112
0925-4005/

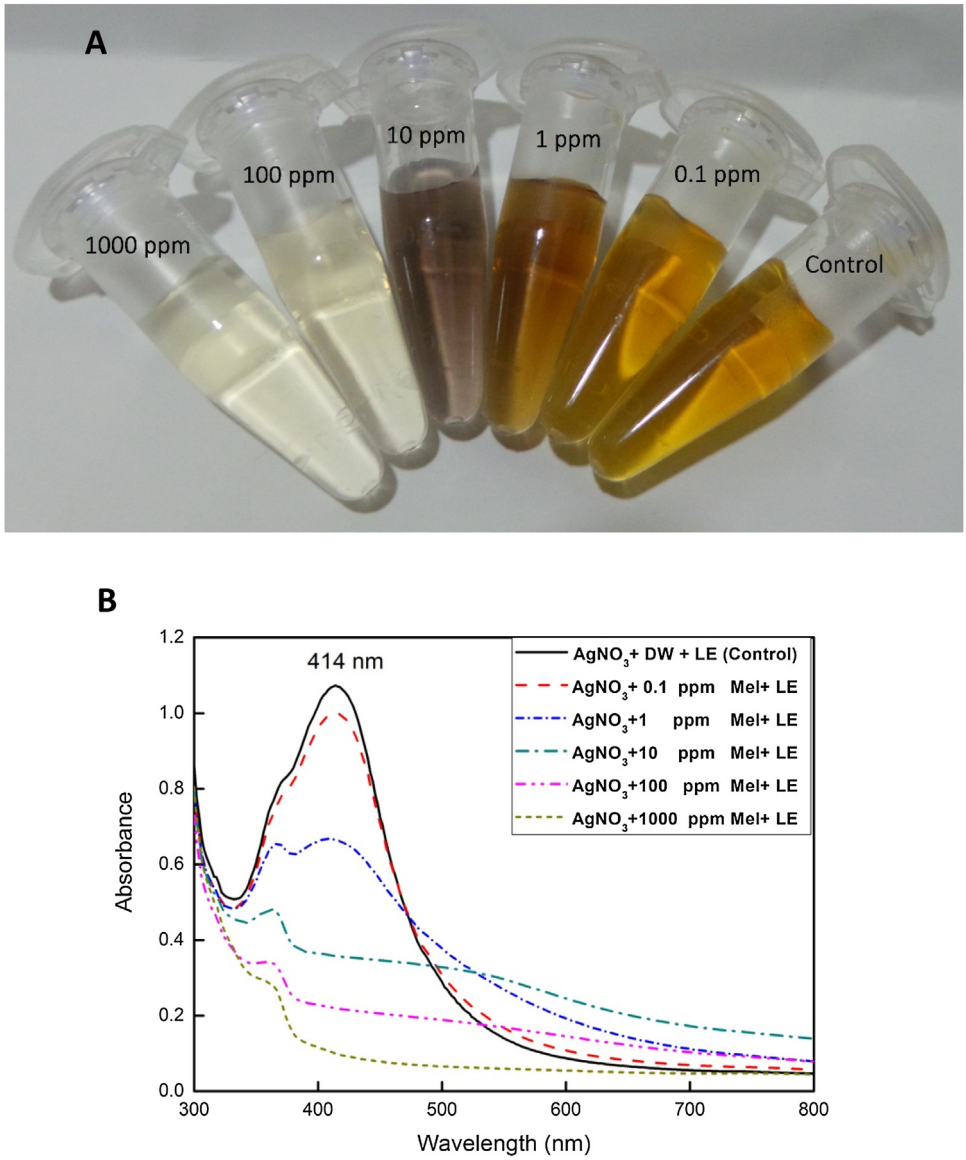

Fig. 1. A) Color photo of sensing of melamine at different ppm concentrations of A –0.1 ppm, 1 ppm, 10 ppm, 100 ppm, 1000 ppm using *Parthenium* sp. leaf extract biosynthesized silver nanoparticles by means of interference biosynthesis. B) UV–vis spectroscopy graph exhibiting the spectral shift in SPR peak of silver nanoparticles synthesized with different ppm concentration of melamine.

sensing of any analyte. This single step synthesis cum sensing of nanoparticles is fast and lead to rapid detection in a few seconds (<20 s). Previous interference based sensing methods use chemicals as reducing agents and the sensing time are in the order of minutes. The current strategy of using interference biosynthesis is a method of biocompatible sensing due to the involvement of leaf extracts as reducing agents instead of chemical reducing agents. Also the present technique is faster (in seconds) compared to existing interference sensing techniques for the detection of melamine. Previously reported sensing strategies employed like synthesis followed by sensing consumes a minimum of 30 min of sensing time without including the synthesis time, where as in the current interference biosynthesis based sensing, sensing along with synthesis happens in seconds leading to faster detection of melamine.

2. Materials and method

Silver nitrate (Merck) is used as such for preparation of 1 mM precursor solution. Leaf extract of *Parthenium histerophorus* (noxious weed) has been prepared [19] by boiling 30 g of leaves of in 100 ml deionized water for one hour. Caffeic acid (Merck) was dissolved in water at appropriate concentration (4 mM) and was also used as a reducing agent for interference sensing. For interference based biosynthesis based sensing experiments, 200 μl of different concentrations of analyte (melamine) is added to 500 μl of the precursor (silver nitrate) and 10 μl of the pH adjusted reducing agent (leaf extract) added subsequently. For synthesis and sensing experiments, previously synthesized nanoparticle solutions added to different concentrations of analytes (0.1 ppm to 1000 ppm of melamine). In all the sensing experiments, the microliter volumes of samples were taken in a 96-well plate and subjected to UV–vis absorption spectroscopy scan from 300 nm to 800 nm using the TECAN 200 INFINITE plate reader. High Resolution Transmission Electron Microscopy analysis was done utilizing a F30 TECNAI HRTEM operating at 200 kV. Sample preparation for HR-TEM was done by adding a drop of the aqueous nanoparticles on a 400 mesh carbon coated copper grid and subsequently dried at 50 °C. Milk spiking is done by adding melamine solutions to 1 ml of raw milk