Talanta 115 (2013) 24–31

Contents lists available at ScienceDirect

Talanta

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

# Biopolymer capped silver nanoparticles as fluorophore for ultrasensitive and selective determination of malathion

N. Vasimalai, S. Abraham John *

*Centre for Nanoscience and Nanotechnology, Department of Chemistry, Gandhigram Rural Institute, Gandhigram, 624302 Dindigul, Tamilnadu, India*

ARTICLE INFO

*Article history:*
Received 12 January 2013
Received in revised form
5 April 2013
Accepted 12 April 2013
Available online 19 April 2013

*Keywords:*
Malathion
Silver nanoparticles
Chitosan
Fluorescence quenching
Pesticide

ABSTRACT

This paper describes a novel luminescent sensor for malathion using chitosan capped silver nanoparticles (Chi-AgNPs) as fluorophore. The Chi-AgNPs were synthesized by the wet-chemical method and were characterized by absorption, fluorescence, HR-TEM, XRD and DLS techniques. The Chi-AgNPs show the absorption maximum at 394 nm and emission maximum at 536 nm. While adding 10 μM malathion, yellow color Chi-AgNPs was changed to brown and the absorbance was decreased along with a redshift. The observed spectral and color changes were mainly due to the aggregation of Chi-AgNPs. This was confirmed by zeta potential, DLS and HR-TEM studies. No significant absorption spectral change was observed for Chi-AgNPs in the presence of less than micromolar concentrations of malathion. However, the emission intensity of Chi-AgNPs was decreased and the emission maximum was shifted toward higher wavelength in the presence of picomolar concentration of malathion. Based on the decrease in emission intensity, the concentration of malathion was determined. The Stern–Volmer constant, Gibbs free energy change, association constant, quantum yield and binding constant were calculated and the quenching mechanism was proposed. The Chi-AgNPs show good selectivity toward the determination of 10 nM malathion in the presence of 1000-fold higher concentrations of common interferents. A good linearity was observed for the emission intensity against $1\times10^{-9}$–$10\times10^{-12}$ M malathion and the detection limit was found to be 94 fM $L^{-1}$ ($S/N=3$). The proposed method was successfully applied to determine malathion in fruits and water samples and the obtained results were validated with HPLC.



## 1. Introduction

The pesticides assisted to extensively reduce crop losses and to improve the yield of crops such as corn, maize, vegetables, potatoes and cotton [1–3]. Worldwide use of pesticides increased terrifically since the 1960s [3]. Although pesticides are beneficial they adversely affect both the environment and human health. Residues of pesticides pollute the soil and water, persist in the crops and enter the food chain and are finally swallowed by human with foodstuffs and water. Further, pesticides were responsible for contributing to biodiversity losses and deterioration of natural habitats [4–6].

Malathion is an organophosphorous pesticide which is extensively applied on crops like banana, citrus fruits, tobacco and on vegetable crops like tomato, beans, brinjal and carrot. It can control the pests like aphids and similar sap-sucking insects, various weevils, small beetles, scale insects and red spider mites [7–9]. Very recently, in one of the southern states of India, Tamilnadu, 5367 people were affected by dengue fever and about 39 were died [10]. Indian government is making a lot of efforts to control the mosquitoes. Malathion is very widely used in urban centers as a fog against mosquitoes by the public health authorities in India [7–10]. Moreover, it is also used to control animal ectoparasite and human body lice [11].

The acute toxicity of malathion to animals is caused by inhibition of acetylcholinesterase (AChE), an important enzyme functioning to hydrolyze the neurotransmitter acetylcholine (ACh) [8]. The inactivation of AChE causes an over accumulation of ACh and thereby disrupts the nerve transmissions [9]. The genotoxic studies indicate that malathion can also induce DNA damage [12]. Malathion has also been shown to modulate oxidative stress and cholinesterase depression in saliva and plasma and increase plasma glucose concentration associated with stimulation of hepatic glycogen phosphorylase and phosphoenolpyruvate carboxykinase in rats [12]. Toxic symptoms resulting from human exposure to malathion, include breathing problems, headache, nausea and dizziness, while high exposure can produce fetal poisoning [13]. Malathion can persist in the human body for at least two generations [10]. It is suspected to cause kidney problems, human birth defects and child leukemia [11]. Due to its extensive application, it finds its way into surface water bodies through agricultural runoff and municipal waste water. Hence, it is

* Corresponding author. Tel.: +91 451 245 2371; fax: +91 451 245 3031.
*E-mail address:* abrajohn@yahoo.co.in (S. Abraham John).

0039-9140/$ - see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.talanta.2013.04.033

necessary to determine the residues of malathion in water and vegetables to protect the human beings and environment from its hazardous effect.

Generally, malathion was determined by high performance liquid chromatography [14], gas chromatography coupled with mass spectrometry [15], voltammetry [16], infra-red spectroscopy [17] and Raman spectrometry [18]. Among these methods, many of them are time consuming and labor-intensive due to the complex pretreatment, require expensive instrumentation and high cost of personnel and are not readily adaptable to on-site detection [14,15,17,18]. Therefore, it is essential to develop a simple, rapid, low cost and field-portable method for the detection of malathion in environmental samples. In recent years, the determination of toxic chemicals by the spectrofluorimetry method has received much attention [19–22] because of its high sensitivity, selectivity, reproducibility, less time consumption and simple experimental conditions [23–25]. Recently, metal nanoparticles based pesticides sensors have been gaining momentum due to their interesting optical, catalytic and electrical properties [4,5,9,25]. Among the different metallic nanoparticles, silver nanoparticles (AgNPs) show excellent optical properties, good conductivity, chemical stability and catalytic activity [26–28]. The AgNPs have been used extensively as an antibacterial agent, food storage, textile coatings and toxic chemicals sensor [26–29].

Chitosan is a degradable biopolymer (Chart 1) and it has several attractive properties including excellent film forming ability, high permeability toward water, good adhesion, non-toxicity and biocompatibility [30–32]. The derivatives of chitosan have been applied for the removal of various heavy metal ions from aqueous solution [33]. Therefore, we have used chitosan as a capping agent for the synthesis of AgNPs in the present study. The present work describes the luminescent sensor for malathion using chitosan capped silver nanoparticles (Chi-AgNPs) as fluorophore. The Chi-AgNPs show the absorption maximum at 394 nm and the emission maximum at 536 nm while exciting at 394 nm. The emission intensity of Chi-AgNPs was decreased and the emission maximum was shifted to higher wavelength while adding picomolar malathion. Based on the decrease in emission intensity, we have determined the concentration of malathion. The lowest detection limit was found to be 94 fM $L^{-1}$. The obtained results were validated with HPLC.

## 2. Experimental

### 2.1. Chemicals

Silver nitrate, malathion and acetic acid were purchased from Sigma-Aldrich. Sodium borohydride ($NaBH_4$) was obtained from Merck (India). Chitosan with its deacetylation degree of 85% was purchased from Pelican Biotech and Chemicals Labs, Kerala (India). All other chemicals used in this investigation were of analytical grade and used directly without further purification. Millipore Milli-Q (18 MΩ cm) water was used in all the experiments.

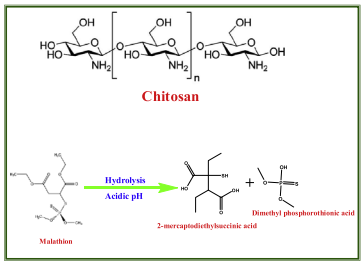


**Chart 1.** Structure of chitosan and hydrolysis of malathion in acidic pH.

### 2.2. Instrumentation

The high resolution transmission electron microscopy (HR-TEM) images of AgNPs were obtained from a JEOL JEM 2100 Advanced Analytical HR-TEM, operating at 200 kV. For TEM measurements, the sample was prepared by dropping 2 μL of a colloidal solution onto a carbon-coated copper grid. UV–visible spectral measurements were performed on a JASCO V-550 UV–visible spectrophotometer. Fluorescence spectra were measured by using a JASCO FP-6500 spectrofluorimeter equipped with a Xenon discharge lamp, and 1 cm quartz cell at room temperature (about at 298 K). A larger volume (500 mL) of Chi-AgNPs was synthesized and the particles were separated by centrifuging at 10,000 rpm and repeatedly washed with water and dried in vacuum. The dried AgNPs powder was used for X-ray diffraction (XRD) analysis. XRD analysis was carried out with a Rigaku X-ray diffraction unit using Ni-filtered Cu Kα ($\lambda = 1.5406$) radiation. Dynamic light scattering (DLS) and zeta potential measurements were performed on Zetasizer Nano S90 (Malvern). Millipore Milli-Q (18 MΩ cm) water was prepared by using Direct-Q Millipore (Cylus Laboratory Equipment, France).

### 2.3. Synthesis of Chi-AgNPs

All glasswares were thoroughly cleaned with freshly prepared aquaregia (3:1; $HCl/HNO_3$) and rinsed comprehensively with Millipore water prior to use. The colloidal solution of chitosan capped AgNPs was prepared as follows. 5 mL of $AgNO_3$ (7 mM) and 1 mL of 1% chitosan in acetic acid were added to 86 mL of water in a round bottom flask with constant stirring. To this solution, 8 mL of 0.05 M $NaBH_4$ was added and the color of the solution turns into yellow immediately after the final addition, indicating the formation of AgNPs. The stirring was continued for another 30 min. The synthesized Chi-AgNPs were purified by centrifugation at 10,000 rpm and then the filtrate was dissolved in Millipore water. The purified Chi-AgNPs were used for further studies.

### 2.4. Real sample analysis

A typical spectrofluorimetric analysis of malathion in polluted lake water and fruit samples was achieved as follows. The mangos and grapes were chosen as the spiked sample to evaluate malathion residues in actual fruits using the Chi-AgNPs. The different concentrations of malathion were sprayed onto mangos and grapes by an atomizer. After 3 days at room temperature, the edible parts of the fruits were taken and crushed well. From the crushed mangos or grapes, 10 g was mixed with 50 mL water in a 100 mL flask and shaken vigorously for 30 min. Then, insoluble draff was removed by a simple filtration. A control sample was also prepared by using the same procedure.

## 3. Results and discussion

### 3.1. Spectral studies of chitosan in the presence of malathion

The UV–visible spectrum of chitosan is shown in Fig. 1a (inset). It shows a shoulder band around 210 nm. Fig. S1 (Supporting information) shows the UV–visible spectra of chitosan in the presence of different concentrations of malathion. The absorbance around 210 nm was decreased after the addition of 100 μM malathion. Further increasing the concentration of malathion, the absorbance around 210 nm was dramatically decreased. No significant decrease in absorbance at 210 nm was observed in the presence of less than 100 μM malathion.