Biosensors and Bioelectronics 64 (2015) 586–593

Contents lists available at ScienceDirect

Biosensors and Bioelectronics

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

Design and fabrication of molecularly imprinted polymer-based potentiometric sensor from the surface modified multiwalled carbon nanotube for the determination of lindane (γ-hexachlorocyclohexane), an organochlorine pesticide

Thayyath S. Anirudhan*, Sheeba Alexander

Department of Chemistry, School of Physical and Mathematical Sciences, University of Kerala, Kariavattom, Trivandrum 695581, India

ARTICLE INFO

Article history:
Received 10 July 2014
Received in revised form
30 August 2014
Accepted 22 September 2014
Available online 30 September 2014

Keywords:
Molecular imprinting
Lindane
Potentiometric sensor
Multiwalled carbon nanotube
Detection limit

ABSTRACT

A novel potentiometric sensor with high selectivity in addition to sensitivity was developed for the determination of lindane, γ-hexachlorocyclohexane (γ-HCCH), based on the modification of γ-HCCH imprinted polymer film onto the surface of Cu electrode. A multiwalled carbon nanotube (MWCNT) was grafted using glycidyl methacrylate (GMA). The reaction of MWCNT with GMA produces MWCNT-g-GMA and the epoxide ring present in the GMA upon reaction with allylamine produces the vinylated MWCNT (MWCNT-CH$=$CH$_2$). MWCNT based imprinted polymer (MWCNT-MIP) was synthesized by means of methacrylic acid (MAA) as the monomer, ethylene glycol dimethacrylate (EGDMA) as the cross linker, α,α′-azobisisobutyronitrile (AIBN) as the initiator and γ-HCCH, an organochlorine pesticide molecule, as the template. The optimizations of operational parameters were also done. Organized material was characterized by means of FTIR, XRD, Raman spectra and TEM analyses. The sensor responds to γ-HCCH in the range 1×10^{-10}–1×10^{-3} M and the detection limit was found to be 1.0×10^{-10} M.

1. Introduction

Lindane, gamma-hexachlorocyclohexane (γ-HCCH), is an organochlorine pesticide, which is used both as an agricultural insecticide (Randenberger Maes and Robert, 1997) and a pharmaceutical treatment for lice and scabies (Agency for Toxic Substances and Disease Registry, 2005; Commission for Environmental Cooperation., 2005). It has an oral LD$_{50}$ of 88 mg/kg in rats and a dermal LD$_{50}$ of 1000 mg/kg. γ-HCCH is highly volatile, and when applied to field crops in particular a major portion (up to 90%) of the pesticide enters the atmosphere and is eventually deposited by rain. According to the World Health Organization (WHO) drinking water guidelines, the maximum allowable limit for γ-HCCH is in the range 0.4–155 ng L^{-1} (Gustavson et al., 1990). Most of the adverse human health effects reported for γ-HCCH have been related to agricultural uses and chronic, occupational exposure of seed-treatment workers (Sellergren, 2000) and they are extremely persistent and cannot be degraded aerobically. High level concentrations of γ-HCCH are normally found in the marine environment and in the North Sea (Lohse et al., 1990). Moreover it is found in ecological niches such as water bodies and in crops, leading to major environmental problems.

Molecular imprinting is a widely accepted method for the isolation of synthetic polymers with specific binding sites for a target molecule. It is generally used for the designing of resources with unusual identification capability towards target molecule (Yilmaz et al., 2000; Haupt, 2001; Hassan et al., 2001). In the past decades, successful syntheses of molecular imprinted polymers have been reported. Some of these materials have potential applications such as catalysis (Polborn and Severin, 1999), solid phase extraction (Rao, 1996), membrane separations (Kimaro et al., 2001) and developing sensors (Alkindy et al., 2002). There are still many imprinted polymeric systems which remain less explored, particularly for the direct recognition and sensing of pesticides (Jenkins et al., 2001; Turkewitsch et al., 1998).

A variety of chemical sensors have been synthesized by using MIPs for molecular recognition followed by appropriate signal transductions based on various physical tools such as capacitance (Tanya et al., 2001), conductometry (Sergeyeva et al., 1999), amperometry (Shoji et al., 2002), voltammetry (Lobo-Castanon et al., 2003), quartz microbalance measurements and spectroscopy (Tatyana et al., 2002). The potentiometric sensors, predominantly MIP based sensors, are highly attractive in the area of chemical sensing applications (Jenkins et al., 2001). The

---

* Corresponding author.
E-mail address: tsani@rediffmail.com (T.S. Anirudhan).

http://dx.doi.org/10.1016/j.bios.2014.09.074
0956-5663/© 2014 Elsevier B.V. All rights reserved.

potentiometric sensors do not require the template molecules to diffuse through the electrode membranes for generation of membrane potentials (Blanco-López et al., 2004) unlike in other sensors based transduction techniques. It also has less response time and high sensitivity and permits inexpensive detection of analytes.

Some of the literature reports show that potentiometric and ion sensitive field effective transistor membrane or films were successfully used for the selective detection of atrazine and triazine herbicides (Agostino et al., 2006; Pogorelova et al., 2002). Recently, for selective monitoring of uranium and phorate molecules (Metilda et al., 1997) an imprinted polymer inclusion membrane (IPIM) based sensor has been developed. Earlier workers (Zhou et al., 2004; Prathish et al., 2007) have employed a potentiometric sensor that can be used for the selective detection of methyl phosphonic acid (MPA), a degradation result of chemical warfare agents.

Carbon nanotubes (CNTs) possess a unique position in the field of material science (Iijima, 1991) because of their intrinsic extraordinary physico-chemical, electrical, thermal and mechanical properties. However, the potential applications of multiwall carbon nanotubes (MWCNTs) are limited due to some inadequacy such as chemical inertness of the graphitic network, poor dispersion and interfacial bonding (Wang et al., 2008). An essential criterion for functionalization of MWCNTs is to efficiently disperse the individual nanotubes and establish a strong chemical affinity or adherence of the nanotubes (covalent or non-covalent) with the surrounding matrix (Spitalskya et al., 2010; Hu et al., 2003). The covalent functionalization in turn incriminates ultra-violet photo-oxidation (Parekha et al., 2006), plasma treatment (Felten et al., 2006), ultra-sonication exposure (Park et al., 2002), microwave irradiation (Vazquez and Prato, 2009), etc. whereas the noncovalent functionalization involves π-stacking interactions (Homenick et al., 2007) like surfactant adsorption (Kang and Taton, 2003) and the use of the p-conjugated macromolecules (Vaisman et al., 2006). Furthermore, the epoxy-functionalization of the nanotubes is vital for its use as a primary polymer matrix in the preparation of advanced composite materials, which is essential for load-bearing applications.

The present work reports the modification of MWCNT with glycidyl methacrylate to form a MWCNT-g-GMA composite. The free radical reaction was initiated by the initiator AIBN. Several studies were conducted on the functionalization of MWCNT and grafting with polymer chains using free radical polymerization (Shaffer and Koziol, 2002; Park et al., 2003; Hwang et al., 2004). The main chemical significance is that the MWCNT can disperse well in the polymer matrix by means of free radical polymerization, such as poly-(methyl methacrylate), polystyrene, etc. The reported results reveal that the free radical initiators can activate MWCNT to open its π-bonds, and MWCNT takes part in the additional polymerization of the monomer molecule. Thus the polymer chains grafted onto the surface of the MWCNT form a strong covalent bonding between MWCNT and polymer matrix. The reason for the choice of GMA as the monomer for grafting is due to its dual functionality. Besides the GMA contains an epoxide group; it is useful for introducing any types of functionalities or side chain elongations. The GMA is also suitable to modify the polarity of MWCNT by opening the epoxide ring (Young-Sun and Soo-Jin, 2010). This MWCNT-g-GMA composite upon further reaction with allylamine forms the MWCNT-g-GMA-CH$=$CH$_2$. The MWCNT-g-GMA-CH$=$CH$_2$ undergoes selective polymerization with γ-HCCH to form a complex matrix by means of a covalent bond on the surface of MWCNT. The prepared MWCNT-MIP was characterized by using different characterization techniques.

2. Experimental

2.1. Reagents

γ-HCCH was purchased from SUPELCO, USA. MWCNTs were obtained from Shenzhen Carbon Nanotechnologies Co. Ltd. Trimethylolpropane trimethacrylate (TRIM), methacrylic acid (MAA), 2,2′-azobisisobutyronitrile (AIBN) and EGDMA were purchased from Sigma Aldrich. GMA was purchased from Fluka (Switzerland). Allylamine was obtained from E. Merk India Ltd.

2.2. Apparatus

The concentration of γ-HCCH was determined spectrophotometrically on a JASCO UV–visible (model V-530, Japan) spectrophotometer. The morphological study of MWCNT-MIPs was performed on a Philips CM12 Transmission Electron Microscope (TEM). The Fourier transform infrared (FTIR) spectra were recorded with a Shimadzu FTIR spectrometer in the wavelength range 800–4000 cm^{-1} using a KBr window at a resolution of 4 cm^{-1}. The X-ray diffraction (XRD) patterns of the adsorbent were recorded using an X′Pert Pro X-ray diffractometer using Cu Kα radiations at a scanning speed of 2°/min and at a wavelength of 1.5406 Å. All pH measurements were carried out on a Systronic (model μ pH system 362)-pH meter (Systronic India Ltd.). Potentiometric response characteristics were studied with a $4^{1/2}$ Digit True RMS Multimeter (MODEL 1085). The linear sweep voltagrams were obtained with a VSP-Potentiostat (Bio-Logic-Science Instruments). Raman spectra were recorded with a micro-Raman spectrometer Lab Ram UV HR, Jobin-Yvon.

2.3. Synthesis of GMA grafted MWCNT

The weight ratio of MWCNTs, TRIM and AIBN was 1:280:0.6. The amount of MWCNT content was 15 wt% with respect to GMA concentration. The whole mixture was stirred and sonicated at 65 °C using a nitrogen atmosphere for 2 h. In the subsequent reaction, the GMA grafted MWCNTs slurry was washed several times with acetone to eliminate the nongrafted GMA and finally filtered with a 0.2 μm Teflon microfiltration cell (Chih-Chun et al., 2012).

2.4. Synthesis of vinyl group functionalized MWCNT

To the mixture of 2.3 g of GMA grafted MWCNT and 100 ml of CH_2Cl_2, 6.9 ml allylamine and 13.7 ml titanium isopropoxide were added. The mixture was stirred under 75 °C for 18 h. The mixture was filtered through a 0.22-μm polycarbonate membrane, washed thoroughly with distilled water and dried to get vinyl group functionalized-MWCNT (MWCNT-CH$=$CH$_2$) (Murruzzu and Reira, 2007).

2.5. Synthesis of γ-HCCH imprinted and non-imprinted polymer materials

MWCNT-CH$=$CH$_2$ was added to the solvent mixture of 60 mL of acetonitrile and 10 mL of toluene in a 500 mL round-bottom flask. After that the mixture was purged with N_2 gas under a constant magnetic stirring. A mixture of γ-HCCH and MAA was prepared and dissolved in 35 mL of N,N dimethylformamide. It was stirred for 30 min to get a compound of template molecule and functional monomer. To that mixture the cross linker EGDMA and initiator AIBN were also added; the reaction was allowed to proceed for 16 h at 70 °C. Ethanol was used to remove template molecules. Using the same method a blank non-imprinted