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Double-imprinted potentiometric sensors based on ligand exchange for the determination of dimethoate

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Abstract−This paper describes a dimethoate imprinted polymeric potentiometric sensor for the determination of dimethoate that was designed by dispersing the dimethoate-imprinted polymer in dibutyl phthalate plasticizer and then embedded in polyvinyl chloride matrix. The method is based on molecularly imprinted polymer (MIP), which can be synthesized by using dimethoate as a template molecule, chitosan-Cd(II), and 3-mercaptopropyl-trimethoxysilane as a functional monomer and TEOS, and ephychlorohydrin as a cross-linking agent. We have studied a method for the selective binding behavior of dimethoate compounds on the surface of imprinted polymers which is prepared using ligand-exchange (LE) (chitosan-Cd(II)) monomer. The MIP-LE-1 and MIP-LE-2 based membranes have shown a detection limit of 4.3×10^{-6} mmolL^{-1} and $3.5.10^{-5}$ mmolL^{-1}, respectively. The sensor has exhibited a response time of 35 min for MIP-LE-1 and 120 min for MIP-LE-2. They are stable for more than three months.

Keywords: Potentiometric Sensor, Dimethoate, Mip, Ion Selective Electrode, Pesticide

INTRODUCTION

Molecular imprinting is a promising technique for the preparation of polymers which possess excellently selective recognition ability due to the shape of molecular recognition cavity and the resulting functional group interactions of the target and monomer molecules. Molecularly imprinted polymers (MIPs) possess specific sized cavities with shapes and chemical functionalities with the selective adsorption and desorption of species of interest. An MIP has unique advantages over natural biological receptors in terms of physical and chemical stability, ease of preparation, low cost and application in harsh environmental conditions. Based on these favorable properties, the MIP has been successfully used in many fields, such as chromatographic separation [1-3], immunoassays [4,5], solid-phase extraction [6], chiral separation [7], pharmaceutical analysis [8], recognition of elements in chemical or biological sensors [9-11] and so on.

Double imprinting methodology has been developed to synthesize novel materials. This new methodology makes use of the recent advances in the controlled synthesis of mesoporous materials and combines them with molecular level imprinting techniques. with crown ether ligands. Double-imprinting concept was applied to the preparation of an organic-inorganic hybrid sorbent for selective separation of ions from aqueous solution [12,13]. Sol-gel molecular imprinting is generally carried out by synthesizing the sol-gel materials in the presence of template molecules [12,13]. Subsequent removal of template molecules resulted in a gel network containing imprinted cavities with similar shapes and sizes or even specific chemical functionalities, corresponding to the original template molecules. The essence of this methodology is the combination of two powerful imprinting techniques at different scales: molecular imprinting and micelle templating synthesis. This produces imprinted mesoporous hybrid sorbent materials that exhibit not only fast binding kinetics and high capacities but also molecular recognition capabilities.

Potentiometry is one of the simplest instrumental techniques that many chemists encounter. Potentiometric sensors provide an exciting and achievable opportunity to perform biomedical, environmental and industrial analyses away from a centralized laboratory because they make it possible to combine the ease of use and portability of potentiometry with simple, inexpensive fabrication techniques. While potentiometry has been used for many years, the advances in the field of ion-selective electrodes make it a valuable technique in the modern laboratory [14,15]. The unique feature of potentiometry with MIP-based sensors is that the species do not have to diffuse through the membrane so there is no size restriction on the template compound. Despite all these advantages, a few MIP-based sensors have been reported that utilize a potentiometric transducer [16-18]. These studies describe potentiometric sensors created in several different ways: by dispersing MIP particles in plasticizer and embedding them in a polyvinyl chloride (PVC) matrix [19-22], by forming a glassy membrane [23], by assembling the template on the polar surface of an indium tin oxide (ITO) glass plate [24,25], by depositing a MIP polymeric film on the gate surface of an ion-sensitive field-effect transistor [26,27] and by embedding MIPs in the carbon paste electrode [28].

We have fabricated a new potentiometric sensor that provides recognizing of dimethoate, with a high selectivity and sensitivity by the use of MIPs as a recognition tool. Organophosphorus insecticides are very toxic compounds, intensively used in agriculture and they irreversibly inhibit the catalytic active sites of Acetylcho-

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linesterase (AChE). Dimethoate is an organophosphorus insecticide. We have proposed novel phosphotriesterase mimick surface imprinted polymeric potentiometric sensor for the selective determination of the nerve agent by chitosan-Cd(II) as a new metal-chelating monomer via metal coordination-chelation interactions and dimethoate (target nevre agent) templates. Dimethoate memories having MIPs have been synthesized using chitosan-Cd(II) and 3-mercaptopropyl-trimethoxysilane as a monomer and tetraethoxysilicate (TEOS) and epichlorohydrin as a cross-linking agent, respectively. We have prepared a dimethoate imprinted polymer based potentiometric sensor by dispersing the dimethoate imprinted polymer particles in dibutyl phthalate (plasticizer), which has been embedded in polyvinyl chloride matrix. This preparation method for potentiometric sensors for charged species has here been applied because it is simple and straightforward.

EXPERIMENTAL

1. Chemicals

Chitosan, dibutyl phthalate (DBP), high relative molecular weight polyvinyl chloride (PVC) and 3-mercaptopropyl-trimethoxysilane were supplied from Aldrich Chemical (USA). Nitric acid, sodium hydroxide were purchased from Merck (Darmstadt, Germany). Paraoxan (PO), dimethoate, 4-aminoantipyrine, and azobisisobutyronitrile (AIBN) were supplied by Aldrich and used as received. Parathion (PT) was supplied from Supelco/Chem Service. Tetraethoxysilicate (TEOS) from Acros Organics (Belgium) and cadmium nitrate monohydrate from Fluka (Poland) were used in this study.

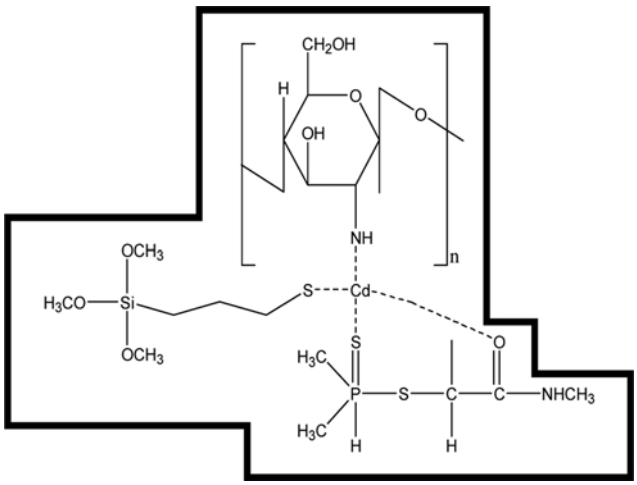
2. Apparatus

Metler Toledo Seven Multi pH-ionmeter was used to measure pH values and ion concentration. Pesticide concentration was measured by unicam UV spectrometer. FTIR spectroscopy was used in the 4,000-400 cm^{-1} range for the chemistry of functional monomer, pre-organized complex, and imprinted beads in the solid state (FTIR 100 series, Perkin Elmer, USA).

3. Preparation of Dimethoate-imprinted Polymer Based on Ligand-exchange

3-1. Dimethoate-double-imprinted Polymer with Chitosan-Cd(II)-3-mercaptopropyltrimethoxysilane (MIP-LE-1)

The functional metal-chelate monomer, chitosan-Cd(II)-3-mercaptopropyl-trimethoxysilane was chosen to interact with dimethoate to make metal-complexing polymeric receptors for selective binding of dimethoate (Schema 1). This preorganization complex defines the size and direction of the chemical interactions of the paraoxon imprinted cavity to prepare synthetic dimethoate. To pre-organize chitosan-Cd(II)-3-mercaptopropyl-trimethoxysilane-dimethoate ligand-exchange monomer, chitosan (6 mmol) was dissolved in the CH_3COOH (5%) and $Cd(NO_3)_2.H_2O$ (3 mmol) was added slowly into this solution with continuous stirring at room temperature. Then, 3 mmol of 3-mercaptopropyl-trimethoxysilane and dimethoate (3 mmol) were added to the mixture. The mixture solution was slowly dropped into 150 mL of 1 M aqueous NaOH. The suspended solution was stirred at 200 rev/min for 12 h. After filtration of the suspended solution and drying in vacuo, the obtained mixture was crosslinked by 3 mL of TEOS in 250 mL of the CH_3COOH (1%) under refluxing conditions in an oil bath (ca. 110 °C) for 8 h.



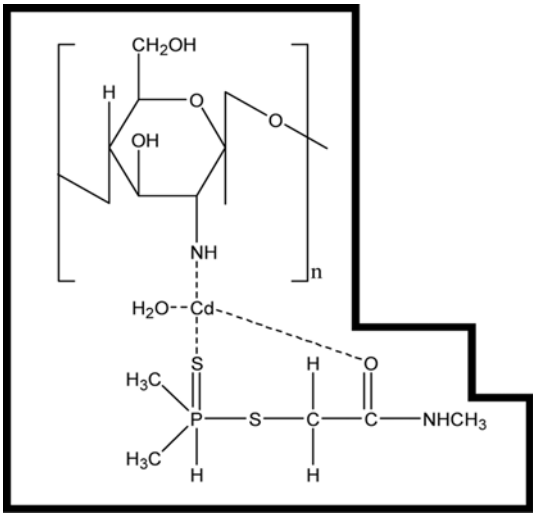
Scheme 1. Creation of recognition sites for dimethoate on MIP-LE-1.

Then, 250 mL of 0.1 M aqueous NaOH solution was added to complete the crosslinking reaction. After vacuum filtration, the product of dimethoate-imprinted polymer was washed with 0.1 M formaldehyde and deionized water several times to remove dimethoate. The resulting product was the dimethoate-imprinted polymer.

FT-IR spectra of MIP-LE-1 are given below: FT-IR (KBr, cm^{-1}): 3,388 cm^{-1} (-OH band), 2,927 cm^{-1} (aliphatic C-H band), 1,568 cm^{-1} (N-H band), 1,043 cm^{-1} (Si-O), 568 cm^{-1} (Cd-S band). After the removing of dimethoate, the dimethoate band at the 568 cm^{-1} was not seen in the IR spectra of MIP-LE-1. This result indicated that dimethoate was connected to chitosan-Cd(II). Thus, it is understood that selective cavities for dimethoate in the MIP-LE-1 have been formed.

3-2. Dimethoate-mono-imprinted Polymer with Chitosan-Cd(II) Ligand-exchange Monomer (MIP-LE-2)

The functional metal-chelate monomer, chitosan-Cd(II), was chosen to interact with dimethoate to make metal-complexing polymeric receptors for selective binding of dimethoate (Scheme 2). This preorganization complex defines the size and direction of the chemical interactions of the paraoxon imprinted cavity to prepare



Scheme 2. Creation of recognition sites for dimethoate on MIP-LE-2.

synthetic dimethoate. To preorganize Chitosan-Cd(II)-dimethoate ligand-exchange monomer, chitosan (6 mmol) was dissolved in the CH_3COOH (5%) and $Cd(NO_3)_2$. H_2O (3 mmol) was added slowly to this solution with continuous stirring at room temperature. And then, dimethoate (3 mmol) was added to the mixture. The mixture solution was slowly dropped into 150 mL of 1 M aqueous NaOH. The suspended solution was stirred at 200 rev/min for 12 h. After filtering this white suspended solution and drying in vacuo the obtained mixture was crosslinked by 5 mL of epichlorohydrin in 250 mL of the acetic acid (5%) under refluxing conditions in an oil bath (ca. 110 °C) for 1 h. Then, 250 mL of 0.1 M aqueous NaOH solution was added to complete the crosslinking reaction. After vacuum filtration, the polymer was washed with 0.1 M formaldehyde and deionized water several times to remove dimethoate.

FT-IR spectra of MIP-LE-2 are given below: FT-IR (KBr, cm^{-1}): 3,417 cm^{-1} (-OH band), 2,378 cm^{-1} (R-S-H band), 2,964 cm^{-1} aliphatic (C-H band), 1,571 cm^{-1} (N-H band), 563 cm^{-1} (Cd-S band). After the removing of dimethoate, the dimethoate band at 563 cm^{-1} was not seen in the IR spectra of MIP-LE-2. This result indicated that dimethoate was connected to chitosan-Cd(II). Thus, it is understood that selective cavities for dimethoate in the MIP-LE-2 have been formed.

4. Preparation of MIP Based Membranes

The PVC membrane electrodes with MIP-LE-1 and MIP-LE-2 were fabricated by following the general procedure. Above-mentioned MIP particles (122 mg), DBP (128 mg) and PVC (100 mg) were added into 2 mL of THF and stirred for 1 h. The obtained mixture was then poured into a glass ring (7.0 cm i.d.) fixed on a glass plate to form a membrane with the thickness of about 0.5 mm. A blank membrane was also prepared in a similar manner, maintaining the same composition, without dimethoate-imprinted particles. The membrane was glued to one end of a Pyrex glass tube with araldite. The tube was then filled with an internal filling solution (mixture of 1 mM KCl and 1mM dimethoate solution).

5. Conditioning of Membranes and emf Measurements

The MIP-LE-1 and MIP-LE-2 based potentiometric sensor was conditioned for 3 days in a 1 mM dimethoate solution and then stirred in formaldehyde for 2 h to remove bound dimethoate ions. The pH-ionmeter was used for the potential measurements at 25.0± 0.1 °C. The measurements were carried out with saturated Ag/AgCl electrode (SCE) as a reference electrode with the following cell assembly.

Cu wire | 1 mM KCl+1 mM dimethoate (internal solution) | (MIP-LE) membrane || test solution | Ag-AgCl, KCl (salt)

6. Characterization of Beads

FTIR spectroscopy was used in the range of 4,000-400 cm^{-1} for the chemistry of mono and double-imprinted polymer in the solid state (FTIR 100 series, Perkin Elmer, USA). The beads (about 0.1 g) were thoroughly mixed with KBr (0.1 g, IR Grade, Merck, Germany), and pressed into a pellet and FTIR spectrum was then recorded.

RESULTS AND DISCUSSION

1. Imprinting Effect

As can be seen in Fig. 1, the potential of the MIP, NIP sensors and control membran increased with the increasing concentration of the

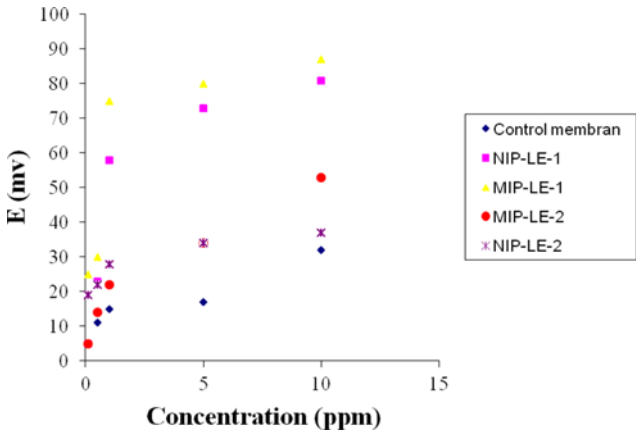


Fig. 1. The imprinting effect on potentiometric sensor (pH 7.0, T: 25 °C).

dimethoate. Dimethoate imprinted microbeads based on ligand, MIP-LE-1 and MIP-LE-2, which were prepared by imprinting technique based on metal-ion coordination interactions, have attractive binding sites and high surface area [29,30], so better accessibility of the active site. In this way, the detection limit decreases and the linear calibration range is extended. As MIP sensor contains dimethoate (template), NIP sensor and control membrane do not contain template molecule. The better characteristic response of MIP based sensor according to NIP and control membrane can be attributed to the imprinting effect.

2. Effect of pH

The pH response profile (Fig. 2) for the MIP-LE-1, NIP-LE-1 and MIP-LE-2, NIP-LE-2 based potentiometric sensors was studied using 10 mmol L^{-1} dimethoate solution in the pH range 6.0-9.0 by adjusting with HNO_3 and NaOH. As can be seen from the figure, the potential remains constant over at pH 7.0, beyond which the potential decreases considerably.

3. Response Time

Response time is yet another important factor that measures the sensing ability of the potentiometric sensor. The response time was recorded at 10 mmolL^{-1} dimethoate solution. The actual potential versus time traces is shown in Fig. 3. The sensor exhibits response

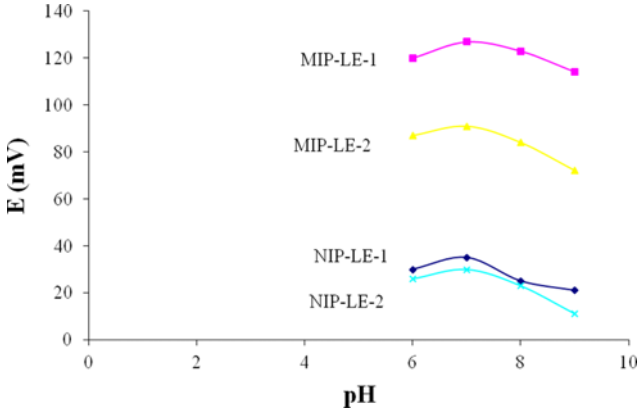


Fig. 2. The influence of the pH on the potentiometric sensor (C: 10 mmol L^{-1}, T: 25 °C).

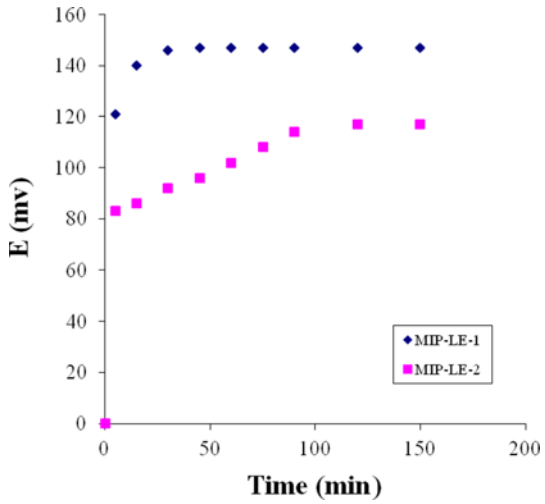


Fig. 3. Responce time of potentiometric sensor (C: 10 mmolL^{-1}, pH 7.0, T: 25 °C).

time of 35 min and 120 min. for MIP-LE-1 and MIP-LE-2, respectively. They can be stable for more than three months.

4. Selectivity

Potentiometric selectivity coefficients, describing the preference of the MIP-LE-1 and MIP-LE-2 based potentiometric sensor for dimethoate (A) relative to an interfering moleule B were determined by the matched potential method, recommended by IUPAC [31]. According to this method, the specified activity (concentration) of the primary ion (A=1×10^{-6} mmolL^{-1}) was added to a reference solution (1.0×10^{-8} mmolL^{-1} of dimethoate) and the potential is measured. In a separate experiment, interfering molecules were added to an identical reference solution and the potential was measured in the same way. The selectivity coefficient was given by the resulting primary ion to interfering molecule activity (concentration) ratio. The selectivity coefficients obtained for MIP-LE-1 and MIP-LE-2 based potentiometric sensors have been compared (see Table 1).

It can be concluded that the dimethoate imprinted particles showed the following ion affinity order under competitive conditions:

for MIP-LE-1,

Dimethoate>Malathion>Parathion>Phosphamidone>Glykophosphate >Paraoxon

for MIP-LE-2,

Dimethoate>Malathion>Glykophosphate>Phosphamidone>Parathion >Paraoxon

Table 1. Comparison of selectivity coefficients of MIP-LE-1 and MIP-LE-2 (pH 7.0, T: 25 °C)

Molecule	Selectivity coefficient (MIP-LE-2)	Selectivity coefficient (MIP-LE-1)
Glykophosphate	1.29.10^{-6}	1.58.10^{-5}
Phosphamidone	1.78.10^{-6}	1.31.10^{-5}
Parathione	4.06.10^{-6}	1.57.10^{-6}
Paraoxon	7.5.10^{-7}	3.31.10^{-7}
Malathion	1.24.10^{-5}	2.39.10^{-5}

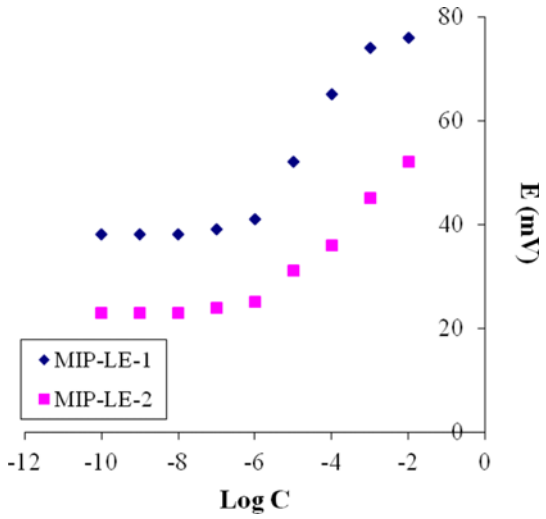


Fig. 4. Potential responses of MIP-LE-1 and MIP-LE-2 membrane based sensors to various dimethoate solutions of increasing concentration (pH 7.0, T: 25 °C).

Note that the imprinted microparticles showed excellent selectivity for the target molecule (dimethoate) due to molecular geometry.

5. Stability, Reusability and Analytical Performance

The important criterion required for chemical sensors in addition to selectivity is stability and reusability. The above developed MIP-LE-1 and MIP-LE-2 potentiometric sensors were found to be stable (deviation is less than 1 mV for 5×10^{-5} mmolL^{-1} of dimethoate) for three months and can be reused for more than 20 times without any loss in sensing ability.

The potential response obtained using the sensors prepared by MIP-LE-1 and MIP-LE-2 is depicted in Fig. 4. The MIP-LE-1 and MIP-LE-2 based membrane showed detection limit of 4.3×10^{-6} mmolL^{-1} and 3.5.10^{-5} mmolL^{-1}, respectively.

Table 2. Comparison of detection limit of reported organophosphate potentiometric sensor

Organophosphate	Detection limit	Life time	Reference
Butyrylcholine	1.8 10^{-7} mol L^{-1}	-	[32]
Butyrylcholine	8×10^{-7} mol L^{-1}	6 month	[33]
Paraoxon, ethyl parathion, methyl parathion	2 μ mol L^{-1}	1 month	[34]
Dimethoate	3.5.10^{-5} mmol L^{-1} for MIP-LE-1 4.3×10^{-6} mmol L^{-1} forMIP-LE-2	3 month	This work

D'Agostino et al. [23] studied the potentiometric sensor for atrazine based on a molecular imprinted membrane that can be synthesized by using phosphate as a template molecule, methacrylic acid as a functional monomer and ethylene glycol dimethacrylate as a cross-linking agent. After that, the template ions (i.e., atrazin) have been removed using methanol/acetic acid solution. Potentiometric sensor based on MIP was described for the determination of atrazine in aqueous solution. Detection limit of molecular imprinted membrane was reported around 2×10^{-5} molL^{-1}.

In our study, detection limits of MIP-LE-1 and MIP-LE-2 membrans were found to be 4.3×10^{-6} mmolL^{-1} and $3.5.10^{-5}$ mmolL^{-1}, respectively. Attractive features of this MIP-LE-1 and MIP-LE-2 based potentiometric sensor are rather low detection limit and its life time for dimethoate.

CONCLUSIONS

Molecule imprinting in synthetic polymers is a process where functional and crosslinking monomers are copolymerized in the presence of target analyte (the imprint, dimethoate in the present case) which acts as an molecular template. The imprint ion is complexed with chitosan-Cd(II) and 3-mercaptopropyl-trimethoxysilane, dissolved in CH_3COOH. Then, the molecular template is held rigidly in a polymeric matrix by polymerizing in the presence of Ephyclorohydrin and TEOS (crosslinking monomer). Subsequent treatment with 1 M formaldehyde removes the imprinted molecule. This situation results in binding sites that are complementary in size and shape to the analyte. Potentiometric technique is a sensitive, simple and inexpensive detection technique; it would be a promising alternative for dimethoate analysis in food samples. In this study, a potentiometric sensor based on MIP was described for the determination of dimethoate in aqueous solution. By using the dimethoate-imprinted polymer (MIP-LE-1 and MIP-LE-2) as the recognition material, a novel dimethoate imprinted sensor with good reproducibility, wide linear range, low detection limit and high selectivity was developed. The detection of dimethoate by the dimethoate imprinted sensor is sensitive in the potentiometric sensor system. The present study highlights the homogeneous dispersion of leached MIP particles in PVC membrane and the study of the potentiometric response which seemed to depend on the ability of MIP to change its conformation upon analyte bonding. As a conclusion, dimethoate imprinted microbeads based on ligand, MIP-LE-1 and MIP-LE-2, which were prepared by imprinting based on metal-ion coordination interactions, have an attractive binding mechanism and high surface area, so better accessibility of the active site.

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