Colorimetric bismuth determination in pharmaceuticals using a xylenol orange sol–gel sensor coupled to a multicommutated flow system

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Abstract

A new sol–gel Bi(III) sensor was developed by incorporating xylenol orange (XO) into sol–gel thin films (<1 µm thick) coated on glass slides. Several sols were produced in order to evaluate the effect of different processing parameters on the final characteristics of the sensor. Sensor films based on tetramethoxysilane (TMOS) as precursor, nitric acid catalysis, water:alkoxide ratio of 2 and XO concentration of 1.5 g l−1 were found to be the most suitable to be used as Bi(III) sensors. They presented good sensitivity, reversibility and stability, low leaching and fast response time in the proposed working conditions. These sensors were coupled to a multicommutated flow system for the determination of Bi(III) in pharmaceutical products. The absorbance of the Bi(III)-immobilized XO complex formed was monitored at 515 nm. The regeneration of the sensor was accomplished by flowing a Cl− ion solution through the flow cell containing the sensor membranes. The procedure enables a relative standard deviation of results better than 0.8%, an analytical concentration range between 125.0 and 875.0 g l−1, a detection limit of 7.0 g l−1 and a sampling frequency of 45 samples per hour. The results obtained on real samples analysis were compared with those obtained by EDTA titrimetric method (British Pharmacopoeia), with relative deviation errors inferior to 5%.

Keywords: Bi(III) optical sensor; Sol–gel films; Multicommutated flow analysis; Pharmaceutical formulations

1. Introduction

The sol–gel technology has been widely used in the preparation of sensors and biosensors since it provides a simple means to incorporate, at low temperature, organic and biological recognizing elements in an optically clean, mechanically and thermally stable inert support [1,2]. Typically, a sol is first formed by mixing a liquid alkoxide precursor, water, a co-solvent (usually ethanol or methanol) and a catalyst (acid or base) at room temperature [3]. Through continuous monomer hydrolysis and condensation reactions, a porous gel network is obtained. Afterwards, gel aging and drying can be conducted under controlled conditions in order to obtain densified solid matrices. During the steps of hydrolysis, condensation or aging, the sensor recognizing elements can be easily added and become entrapped in the support net, yet remaining sterically accessible to small analytes that diffuse into the pore network. A major advantage of sol–gel technology results from the process ability to tailor the network structure, thickness, pore size, and pore distribution by appropriate control of the variables of the preparation procedure [4,5].

When sensor optical transduction is intended flow cells using wave-guide coated sol–gel sensor films are commonly resorted due to their short response time and good optical transparency. By coupling sensing devices to automated continuous flow techniques mutual advantages can be observed [6]. First, simpler manipulation of solutions, simplified manifolds, reagent economy, and cleaner determination procedures are feasible. Moreover, sensor performance characteristics such as selectivity improvement, reduced response time, suitable regeneration, and stable extended use of sensors are obtained through their use as detectors in flow manifolds. This is the case of sensors towards metal ions, in which adequate selectivity is difficult to attain [7]. This paper reports on the development of a new Bi(III) optical sensor based on the immobilization of xylenol orange (XO) in a sol–gel thin film. The sensor was included...
Bismuth compounds have been used orally for their anti-infective and anti-diarrheal properties in a wide range of gastrointestinal tract disorders. Certain salts have antibacterial effects against *H. pylori* and are used particularly for the treatment of peptic ulceration and peptic gastritis. Bismuth has also been applied topically in skin disorders and anorectal disorders such as haemorrhoids [10]. Although bismuth intestinal absorption is low, toxicity is possible with excessive overdosage and is related with neurotoxicity, encephalopathy, and kidney damage [10,11]. Numerous procedures have been reported for the assessment of bismuth in pharmaceutical products. Official methods are based on EDTA titration with several indicators; xylenol orange is used in the current British Pharmacopoeia method [12]. Several spectrophotometric methods have been reported [13–22] and some of these include flow injection procedures [15,17,21,22]. Most of them are based on previous liquid–liquid extraction procedures [13,14,16–21], which are time consuming and require controlled reaction conditions. The method proposed in the present work is based on the reaction of Bi(III) with immobilized XO in strongly acidic medium to form a red colored XO–Bi(III) complex which is monitored colorimetrically at 515 nm. Cl− ion, which forms stable complexes with Bi(III), is used for regeneration of the sensors.

2. Experimental

2.1. Reagents and solutions

Analytical grade chemicals were used without further purification. Milli-Q (Millipore) de-ionized water was used to prepare all solutions.

Tetramethoxysilane (TMOS, product no. 87680); methyltrihydroxysilane (MTES, product no. 69435); polyethylene glycol 6000 (PEG, product no. 81253); and nitric acid (product no. 84385) were from Fluka. Xylenol orange tetrasodium salt (product no. 108677), ethanol, sodium chloride, chloridric acid, and sulphuric acid were purchased from Merck.

A stock standard solution containing 1.00 g l−1 Bi(III) was prepared by dissolving 0.2321 g of bismuth(III) nitrate pentahydrate (product no. 24,859-2, Aldrich) in 50.0 ml HNO₃. The solution was diluted again in 100.0 ml of 7 mol l−1 HNO₃.

Four pharmaceutical products available in the Portuguese market (Pylorid™, Gastrimuto® and De-Nol® tablets and Synalar® Rectal ointment) were used as samples. Five tablets of each tablet sample were weighed, ground in a mortar and homogenized. From each powdered sample and from the ointment an accurately weighed amount was taken (corresponding to ca. 5.0 mg of bismuth) and 4.0 ml of 14.0 mol l−1 HNO₃ were added. The mixture obtained was evaporated nearly to dryness on a sand-bath [22]. This procedure was repeated three times to ensure complete removal of possibly existing Cl− ions in the form of volatile HCl. The residue was dissolved in 100.0 ml of 7.0 mol l−1 HNO₃ and filtered if necessary. From this solution, 2.0 ml were taken and dissolved again in 100.0 ml of 7.0 mol l−1 HNO₃.

2.2. Apparatus

Conventional absorbance measurements for sensors characterization were performed in a Lambda 45 UV-Vis spectrophotometer (Perkin-Elmer, Shelton, CT, USA). Thickness of the sol–gel films was measured in an Alpha-Step 200 profilometer (KLA/Tencor Instruments, Milpitas, CA).

The flow manifold comprised four NResearch 161 T031 three-way solenoid valves (Stow, MA, USA) and a Gilson Minipuls 3 peristaltic pump (Villiers-le-Bel, France) equipped with isoviscous pump tube of the same brand. Flow lines were built using PTFE 0.8 mm i.d. tubing. For absorbance measurements in the flow system plastic optical fibers (POF1-ST) connected to a digital color wheel source (FOCWDIG), a silicon photodiode detector (VISD), and a photodetector amplifier (PDA1) from World Precision Instruments (WPI, FL, USA) were used. The manifold devices control and signal acquisition were accomplished through a home-made QuickBasic 4.5 software program, using a computer equipped with a PCL 711 S Advantech interface card.

2.3. Preparation of sol–gel thin films

Square glass slides (18 mm x 18 mm) were used as substrates for film deposition. Prior to film coating, the substrates were treated with concentrated nitric acid, ethanol, and rinsed with distilled water, followed by drying at 100 °C, in order to activate the silanol groups on the surface of the glass. Ten different types of sol–gel thin films were prepared to evaluate the effect of different reaction parameters on the final characteristics of the sensor. Water:alkoxide ratio, pH, type of acid used as catalyst and type of precursor (TEOS, TMOS, and MTES), concentration of immobilized XO and the addition of a surfactant (PEG 6000) were studied. The compositions of the sols, their aging times, as well as the concentration of XO and the molar ratio of acid (H₂SO₄ or HNO₃) to alkoxide precursor (R) are summarized in Table 1. All the sol–gel solutions were prepared in Teflon vials and allowed to gelate at room temperature. They were mechanically stirred during all aging process prior to coating. Films 1–8 were deposited by dip-coating (3 cm min⁻¹); films 9 and 10 were obtained by spin-coating (80 µl of solution coated at 3000 rpm for 15 s). After coating, all films were dried at room temperature for several days.

2.4. Procedures

First of all, triplicate thickness measurements for each type of film were obtained by profilometry in order to
Table 1

Composition of sols for preparation of sensor thin films

<table>
<thead>
<tr>
<th></th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
<th>F4</th>
<th>F5</th>
<th>F6</th>
<th>F7</th>
<th>F8</th>
<th>F9</th>
<th>F10</th>
</tr>
</thead>
<tbody>
<tr>
<td>TMOS (ml)</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>4.0</td>
<td>–</td>
</tr>
<tr>
<td>TEOS (ml)</td>
<td>3.6</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
<td>4.0</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>MTES (ml)</td>
<td>9.0</td>
<td>0.4</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Ethanol (ml)</td>
<td>9.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
<td>16.0</td>
</tr>
<tr>
<td>XO (mg)</td>
<td>18.00</td>
<td>30.97</td>
<td>30.97</td>
<td>30.97</td>
<td>20.65</td>
<td>41.30</td>
<td>61.95</td>
<td>30.97</td>
<td>31.47</td>
<td>31.94</td>
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<tr>
<td>HCl (10⁻² mol l⁻¹) (ml)</td>
<td>–</td>
<td>0.648</td>
<td>–</td>
<td>–</td>
<td>0.645</td>
<td>0.645</td>
<td>0.645</td>
<td>–</td>
<td>–</td>
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</tr>
<tr>
<td>PEG + HCl (10⁻² mol l⁻¹) (ml)</td>
<td>–</td>
<td>–</td>
<td>0.645</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Concentrated HCl (ml)</td>
<td>0.180</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>HNO₃ (10⁻² mol l⁻¹) (ml)</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>0.645</td>
<td>0.980</td>
<td>1.290</td>
<td></td>
</tr>
<tr>
<td>H₂SO₄ (0.05 mol l⁻¹) (ml)</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>0.645</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>XO (g l⁻¹)</td>
<td>1</td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>Gelation time (h)</td>
<td>24</td>
<td>&gt;24</td>
<td>22</td>
<td>48</td>
<td>20</td>
<td>&lt;20</td>
<td>&lt;20</td>
<td>20</td>
<td>24</td>
<td>24</td>
</tr>
</tbody>
</table>

* Aging time of the solution before coating.

evaluate the influence of coating method and aging time of sol-gel solution. To enable immobilized XO and its complex with Bi(III) spectra, solutions containing between 0.1 and 0.5 mg l⁻¹ in Bi(III) were prepared in increasing concentrations of H₂SO₄ and HNO₃ solutions. Afterwards, the films were immersed in each solution and the corresponding spectra were recorded in the wavelength range of 400–600 nm against a bare glass slide immersed in the same solutions. The extent of leaching was evaluated by immersing up to 60 times each prepared sol-gel film doped with XO for 60 s intervals in 2.0 ml of 7.0 mol l⁻¹ HNO₃ solution. After each interval the sensor was withdrawn and the absorbance of the solution registered at the wavelength of 440 nm (wavelength of maximum absorbance for XO). The response time of the sensor films and their analytical performance were accomplished at the wavelength of 515 nm (maximum absorbance for the XO-Bi(III) complex) under flow conditions by incorporating two slides of each type in a home-made flow through cell with an internal volume of 90 μl (Fig. 1a) by coupling it to a multicommutated flow system (Fig. 1b).

The flow manifold contained four three-way solenoid valves, V₁, V₂, V₃, and V₄, which enabled the access of sample, carrier, or regeneration solution by a programmed activation cycle (Fig 1c). The substitution of solutions accessing the system through valves V₁ (sample), V₂ (carrier) and V₃ (regeneration solution) was made possible by simultaneous activation of each one of these valves together with valve V₁ during 10 s. The sample flow through the mixing coil L placed before the detector was ensured by simultaneous activation of valves V₁ and V₂ for 40 s. During this period 10 cycles ON/Off lasting 4 s each were applied to each valve. By this way a binary sampling of sample and carrier (HNO₃ 7.0 mol l⁻¹ solution) allowed the insertion of small plugs of each solution, which after mixing in coil L enabled the appropriate conditions for bismuth(III) sensing. By selecting different ON/Off time ratios different sample/carrier dilutions could be produced. This procedure was used for calibration purposes resorting to only one 1000.0 μg l⁻¹ bismuth calibrating solution. To achieve sensor regeneration 10 ON/Off cycles were applied simultaneously to valves V₂ and V₃. In the same manner as for sample flow, small plugs of 0.5 mol l⁻¹ sodium chloride solution were inserted between small plugs of carrier solution.

3. Results and discussion

3.1. Optimization of sol-gel films preparation

Xylenol orange is widely used as a photometric reagent in the determination of several cationic species including bismuth, yielding reddish violet complexes [23]. It has also
been used in sol–gel sensors to accomplish lead determinations [24]. Hence, this reagent was selected for implementation of the bismuth sensor. Considering the goals of a good sensor membrane, namely high selectivity, reversibility, fast response, and stability, several types of sol–gel membranes were prepared to evaluate the influence of the type of alkoxide monomer, water:alkoxide ratio, nature and concentration of catalyst, XO concentration, aging time, and coating method (Table 1). Therefore, the most frequently used precursors in the sol–gel process, tetramethoxysilane and tetraethoxysilane, were used, being an amount of twenty coated slides of each type prepared for testing and mean results evaluated. It was decided to include also methyltriethoxysilane once previous reports evidenced that it could influence porosity and contribute to stabilize the films [3,25]. All the TEOS- or TMOS-based films prepared presented a yellow color with absorption maximum around the wavelength of 440 nm. The spectra obtained were similar to that obtained for XO in acidic solution, evidencing that the immobilization procedure does not affect significantly its chemical structure. However, for films containing MTES a hypsochromic effect of about 7 nm was registered, evidencing a perturbation of XO electronic structure (Fig. 2).

When MTES was used as single precursor (F1) the films obtained had very low optical transparency, which made them inadequate to be used as optical sensors. The co-polymerization of MTES with TEOS (F2) rendered cloudy films, still with tolerable optical quality, but this strategy proved to be ineffective for the production of this particular sensor. As can be seen in Fig. 3, films F2 showed initial high leaching rate, which, despite being inferior to similar films based on TEOS alone (F5), was still high when compared to the remaining films. In addition, for films F2 no colorimetric response in the presence of bismuth was registered after the leaching trials, suggesting that final porosity was insufficient for analyte diffusion in the membrane or that their hydrophobic characteristics impair their analytical application. In aqueous media TMOS has a larger hydrolysis rate when compared with TEOS, and so the gel point was reached faster and the films obtained had decreased porosity, which was confirmed for membrane F9, for which no significant leaching was registered even after long immersion periods in acidic solution (Fig. 3). As an alternative to the use of MTES concerning film homogeneity and leaching profile, the co-entrapment of PEG 6000 was tried (F3) [3,26]. As a result, reduced leaching was observed (Fig. 3), but also reduced sensitivity and increased response times (Fig. 4) were obtained when films F3 were applied to bismuth determination.

Chloridric (F1–F3, F5–F7), nitric (F8–F10), and sulfuric (F4) solutions were tested as catalysts. All the films obtained presented uniform aspect, but the use of chloridric acid corresponded to films with higher leaching characteristics when compared with those obtained with nitric acid. The use of sulfuric acid as catalyst showed to be inadequate once the sol needed at least 48 h of gelation, and since it speeds up
the hydrolysis rate of ethoxy groups it allows the formation of peculiar particles which impairs further condensation after the coating step [27]. Thus, films F4 became disrupted when immersed in acidic media containing the analyte. Films F5–F7 were prepared with increasing concentrations of xylenol orange. For the sensors prepared with 1.0 g l\(^{-1}\) in XO (F5) the developed color was very tenuous. The use of XO in concentrations higher than 2.0 g l\(^{-1}\) rendered films with irregular surface and low optical transparency. As a compromise, the concentration of 1.5 g l\(^{-1}\) in XO was fixed in the remaining studies.

The increase of molar water/alkoxide ratio (\(R\)) corresponds to a decrease in thickness, shrinkage, and pore volume of sol–gel films, which is consistent with faster hydrolysis and condensation rates due to increased water content [28,29]. Under the conditions of low \(R\) value (\(R = 2\), F8 and F9) a matrix with a more open structure is produced, which seems to be favorable for the formation of the Bi(III)–XO complex. Furthermore, increased sensitivity was obtained. With higher \(R\) (\(R = 4\), F10), films presented low leaching profile (Fig. 3) but they lost analytical response after the leaching trials.

Having established the optimized composition of XO sol–gel sensor films, the influence of the coating procedure was evaluated by resorting to dip-coating and spin-coating techniques. Withdrawal speed of the glass slide from the sol–gel solution in dip-coating procedure, and spin rate, time and coating solution volume in spin-coating procedure were optimized by a trial and error basis in order to obtain smooth and homogeneous coatings. In these conditions a mean film thickness of about 1.0 ± 0.1 μm for dip-coating procedure and of about 0.46 ± 0.02 μm for spin-coating were obtained. However, sensors obtained by dip-coating exhibited increased response time due to their enlarged thickness (Fig. 4) albeit equal sensitivity. Hence, it was decided to proceed the application to real analysis considering only the sensors obtained by spin-coating (F9).

### 3.2. Sensor performance under flow conditions

In solution, the complex formed between XO and Bi(III) presents an absorbance maximum between 540 and 545 nm if sulfuric acid or nitric acid solutions with concentrations in the range 0.05–0.1 mol l\(^{-1}\) and 0.08–0.12 mol l\(^{-1}\), respectively, are used [23,30,31]. To evaluate the developed sensor under flow conditions two of the coated slides were inserted in a home-made flow-cell (Fig. 1a), which was coupled to a multicommutated flow system (Fig. 1b). This was selected once its binary sampling characteristics enable homogeneous flow streams of reactants, which are important when minimization of Schlieren noise is intended [8,32]. Therefore, flow-rate was fixed at 0.8 ml min\(^{-1}\), and equal and reduced activation times of valves \(V_1\) or \(V_3\) were selected in order to provide appropriate mixture of sample (or chloride solution) with the carrier (nitric acid solution). Different chemical conditions were observed for color development in the sensing films. For all the prepared films (F1–F10) the complex XO–Bi(III) exhibited maximal absorption between 515 and 517 nm (Fig. 2) and the reaction only occurred when nitric acid solution with concentration above 4.0 mol l\(^{-1}\) was used as carrier. This non-classical behavior evidenced by trapped XO could result from two factors: (i) the complex equilibrium of XO–Bi(III) binding [31] and (ii) a modification of XO induced by the heterogeneous microenvironment in which the molecule is immobilized [33–36]. Xylenol orange possesses two independent chelating groups and so it can react with Bi(III) to yield different coordinated chelates. The complexes formed over the pH interval from 0.2 to 1.0 have a Bi : XO = 1 : 1 composition and absorption maximum at 550 nm, while at pH above 1.0 complexes Bi : XO = 1 : 2 are preferably formed, with maximum absorption at 500 nm [31]. This strong influence of pH on the type of complex formed is critical when XO is confined in the sol–gel matrix porous network. Several reaction products coexist in the sensor membrane, with different stoichiometry, and probably different formation constants and molar absorptivities. Moreover, a fraction of immobilized XO can remain sterically inaccessible to Bi(III) ions. In this manner, the thermodynamics of complex formation in gel pores may differ considerably from that in bulk solution [24,37].

The second factor is that optical properties of indicator molecules in a sol–gel matrix can change with respect to those of liquid solutions due to physical and/or chemical interactions with the silica rigid cage [33]. The evolution of polycondensation reactions induces changes in the sol–gel matrix that may cause \(pK_a\) shifts in the trapped indicator, directly dependent on the matrix polarity, and change its form [26,34–36]. Pore size and pore-walls characteristics have great influence on the ability of the entrapped ligand to move or reorient inside the pores. Small pores may restrict translational freedom; interactions between surface hydroxyl groups and water modify solvent structure, which affects enthalpy and entropy terms in complexation reactions [34,37].

Since XO-Bi(III) complex is masked by Cl\(^-\), Br\(^-\), and I\(^-\) ions does not affect XO complexes with Zr, Hf, Fe, and Sn, its specific masking behavior may be used to determine Bi(III) selectively in the presence of other metals.
which also form colored complexes with XO [23,30]. Approximately 16 mM of Cl\(^-\) are required to completely mask 100 µg of bismuth [30]. Taking into account that this solution is two-fold diluted with the carrier, a 0.5 mol l\(^-1\) NaCl solution was used for regeneration of the sensors. Under the before mentioned chemical and hydrodynamic conditions, the sensor started to produce response just 17 s after activation of valve V1 (Fig. 4). By injecting a standard solution containing 500.0 µg l\(^-1\) Bi(III) a steady state signal was achieved in 40 s, which underlines the fast response of the developed sensor. The regeneration step was complete 15 s after activation of valve V3.

Five successive calibrations were performed by injection of seven calibrating solutions with concentrations ranging between 125.0 and 875.0 µg l\(^-1\). The general calibration equation (mAU = 0.0345 (±0.5 × 10\(^{-4}\)) × [Bi(III)] + 5.1 (±0.3)) was obtained with a correlation coefficient of 0.9995. The detection limit of 7.0 µg l\(^-1\) was also calculated from the standard deviation of the signals obtained by injection of a blank without analyte (3σ).

A maximum sampling frequency of 45 samples per hour was achieved. The useful lifetime of the developed sensor was also evaluated by consecutive injections of a calibrating solution containing 500.0 µg l\(^-1\) Bi(III), having noticed a decrease of sensitivity after more or less two hundred determinations (Fig. 5).

### 3.3. Real sample analysis

To assess the usefulness of the sensor-based procedure, four pharmaceutical products available in the Portuguese market (Pylorid\(^{TM}\), Gastrimuto\(^{®}\) and De-Nol\(^{®}\) tablets, and Synalar\(^{®}\) Rectal ointment) were selected. The most important potential interference comes from Cl\(^-\) ions present in samples, which form stable complexes with Bi(III). By this, sample pre-treatment was employed to eliminate chloride interference. The possible influence of other ions likely to be present in pharmaceutical samples and also likely to establish stable complexes with XO was checked for the determination of 500.0 µg l\(^-1\) Bi(III). The results obtained (Table 2) evidenced the good selectivity of the developed sensor. The results obtained for the samples examined (Table 3) with five replicate injections (n = 5) are in good agreement with the labeled information given by the manufacturer and with the results obtained with EDTA titrimetric method [12].

<table>
<thead>
<tr>
<th>Ion</th>
<th>Ratio to Bi (w/w)</th>
<th>Change in absorbance (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ca</td>
<td>0.4</td>
<td>-0.4</td>
</tr>
<tr>
<td>Mg</td>
<td>3.5</td>
<td>-3.5</td>
</tr>
<tr>
<td>Zn</td>
<td>1.5</td>
<td>-1.5</td>
</tr>
<tr>
<td>Fe(III)</td>
<td>4.1</td>
<td>-4.1</td>
</tr>
<tr>
<td>Fe(II)</td>
<td>2.0</td>
<td>+2.0</td>
</tr>
<tr>
<td>Pb</td>
<td>4.5</td>
<td>-4.5</td>
</tr>
</tbody>
</table>

Table 2: Effect of potentially interfering ions in the determination of 500.0 µg l\(^-1\) of bismuth

<table>
<thead>
<tr>
<th>Sample</th>
<th>Labeled bismuth content (mg)</th>
<th>Bismuth found (mg)a</th>
<th>EDTA titrimetric method [12]</th>
<th>Bismuth found (mg)b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pylorid(^{TM})</td>
<td>400b</td>
<td>387.2 ± 2.2</td>
<td>387.3 ± 3.9</td>
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<tr>
<td>Gastrimuto(^{®})</td>
<td>400b</td>
<td>387.6 ± 2.3</td>
<td>388.3 ± 3.8</td>
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</tr>
<tr>
<td>De-Nol(^{®})</td>
<td>120b</td>
<td>118.2 ± 0.2</td>
<td>118.4 ± 1.9</td>
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<tr>
<td>Synalar(^{®}) Rectal</td>
<td>50b</td>
<td>50.3 ± 1.6</td>
<td>50.4 ± 0.4</td>
<td></td>
</tr>
</tbody>
</table>

a Mean of three results.
b Mean of five results.
c Milligram of ranitidine bismuth citrate per tablet.
d Milligram of Bi(III) per tablet.
e Milligram of bismuth subgallate per gram of ointment.
4. Conclusions

The sol–gel entrapment of xylene orange provided optical sensing films selective and sensitive to bismuth(III), with low leaching characteristics, fast response, and long life time. Their insertion in continuous flow-systems offers interesting features, such as low detection limit, reagent economy, flow simplicity, and minimum previous pre-treatment. It also proved to be convenient for the determination of bismuth(III) in a variety of pharmaceutical products.

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