RESEARCH PAPER

Gold Nanoparticles Grafted Manganese Doped Fe$_3$O$_4$ Nanoparticles for Determination of Short-Term Release of Silver and Lead from Dental Amalgam in Saliva

Mitra Amoli Diva[1,*], Kamyar Pourghazi[2], Majid Amoli Diva[3]

[1] Faculty of Physics, Sharif University of Technology, Tehran, Iran

[2] Department of Chemistry, Karazmi (Tarbiat Moalem) University, Tehran, Iran

[3] Faculty of Dentistry, Babol University of Medical Sciences, Babol, Iran

ARTICLE INFO

ABSTRACT

Ethylene glycol bisthioglycolate modified gold nanoparticles grafted Mn doped Fe$_3$O$_4$ nanoparticles (Au@Mn-Fe$_3$O$_4$ NPs) adsorbent was synthesized and applied for extraction and preconcentration of trace amounts of Ag$^+$ and Pb^{2+} ions in artificial and natural saliva. The adsorbent was characterized by transmission electron microscopy (TEM), Brunauer–Emmett–Teller analysis (BET), X-ray diffraction spectroscopy (XRD) and vibrating-sample magnetometer (VSM) techniques and Fusayama artificial saliva was prepared and used as a blank sample. Natural saliva samples were collected from nine volunteers who exposed to posterior decayed teeth amalgam filling, and short-term release of heavy metal ions was assessed in 24, 72 and 96 h after filling. The main factors affecting extraction and desorption efficiency of target ions have been investigated. In Optimum conditions, the detection limits of 0.23 and 0.11 ng mL^{-1} with preconcentration factors of 94 and 95 were obtained for Ag$^+$ and Pb^{2+} ions, respectively. The results revealed that the adsorbent has high capacity and good reusability for extraction and preconcentration of target metal ions in relatively high saline solution like biological fluids.

INTRODUCTION

Filling materials especially amalgam are the most frequently used restorative material for dental treatment and many concerns have been raised about them because of the existence of heavy metal ions like mercury, silver, copper, zinc and lead [1]. It was demonstrated that amalgam filling is able to release these metal ions into the oral cavity causing very destructive effects on human body [2, 3]. For example, it was shown that Hg^{2+} release in saliva can represent a continuous source of oxidative damage to oral tissues [4]. It also induces free radical production and changes the redox potential of cells. The release behavior of metal ions in the oral cavity is complex and involves several parameters, such as amalgam type, pH and oxygen variations in the oral cavity, presence of proteins, etc. Hence, certain trends might be outlined based on in vitro determination, and release measurement during laboratory tests should simulate the clinical situations. On the other hand, successful measurement of release through saliva sampling is difficult because of the fast flow of saliva in the oral cavity. In establishing in-vitro tests, synthetic environments are more frequently used than natural body fluids due to

* Corresponding Author Email: *mitraamoli@gmail.com*

easy preparation to artificial saliva solutions. Saliva contains organic and inorganic substances suspended in an aqueous medium. Its pH may vary between 2 to 11 depending on the age, eating habits, and oral hygiene [5]. Thus, metal release from materials used for dental applications has usually been studied in artificial saliva. Thus, artificial saliva solutions have been formulated to fulfilled different requirements. In all of these solutions, strong electrolytes usually NaCl and/or KCl, are present as the main constitute. Furthermore, there exists a buffer system such as HCO$_3^-$/CO$_3^{2-}$ or H$_2$PO$_4^-$/HPO$_4^{2-}$. In some cases, a complex anion like SCN$^-$ is also present which is actually present in human saliva at quite high concentration [6].

Recently, nanotechnology has been intensively applied in different research fields, such as separation science [7], drug delivery [8], and magnetic resonance imaging [9]. Wide application of nanoparticles (NPs) has led to focusing on the use of these nanoparticles in many scientific areas because they have a high surface area which provides high interfacial area and therefore mass transfer is quicker, and equilibrium is achieved more rapidly. An important group of NPs in analytical chemistry have a permanent magnetism (ferromagnetism) or can be attracted by a magnetic field without retaining residual magnetism after the magnetic field is eliminated (superparamagnetism). Both types of magnetic NPs (MNPs) have been successfully used as solid-phase extraction (SPE) sorbents, because they aid the extraction procedure and save time in sample preparation. In conventional SPE, the sorbents are packed into cartridges, disks, or pipette tips, whereas in magnetic SPE (MSPE), which can be also classified as dispersive-SPE (d-SPE), the sorbent is added directly to the sample. After analyte adsorption, MNPs can retain on one side of the extraction recipient with an appropriate supermagnet. For elution, a suitable solvent is added, and the adsorbent is dispersed and retained with the supermagnet [10, 11]. This mechanism facilitates their application in different research areas [12-15]. High affinity to the analyte and reusability of nano-adsorbents provide significant cost reduction for any MSPE procedure. Furthermore, ability to remove adsorbed analytes from the adsorbent is a critical parameter when recovery of metal ions (and not removal) is desired for applications which recycling and resource recovery are needed. These desired features inspire searching a method which has capability of tuning analyte-affinity through direct

adsorbent modification and to obtain an efficient adsorbent, particularly for applications in harsher matrices such as highly saline and biological fluids or processes where repetitive utilization is desired. Known properties of Fe$_3$O$_4$ nanoparticles (NPs) along with good adsorbent characteristics of MnO$_2$ have suggested a hybrid nanomaterial of manganese (Mn) and Fe$_3$O$_4$ NPs which yields a magnetically active adsorbent compare with or better than each precursor [16-19].

In this study, ethylene glycol bisthioglycolate modified gold nanoparticles grated Mn doped Fe$_3$O$_4$ nanoparticles (Au@Mn-Fe$_3$O$_4$ NPs) are used as a magnetic adsorbent for extraction and preconcentration of metal ions in artificial and natural saliva. The level of each target ion is determined by inductively coupled plasma-optical emission spectrometry (ICP-OES). The factors affecting extraction efficiency of the analytes are investigated and optimized. Finally, the proposed method is successfully applied to the extraction and preconcentration of target ions from natural saliva samples.

EXPERIMENTALS

Chemicals and reagents

Lead nitrate, silver nitrate, ferrous chloride tetrahydrate (FeCl$_2$.4H$_2$O), ferric chloride hexahydrate (FeCl$_3$.6H$_2$O), manganese sulphate (MnSO$_4$), hydrogen tetrachloroaurate (HAuCl$_4$), sodium chloride, potassium chloride, calcium chloride (CaCl$_2$.2H$_2$O), sodium dihydrogen phosphate dihydrate (NaH$_2$PO$_4$.2H$_2$O), sodium sulfide nine hydrate (Na$_2$S.9H$_2$O), urea (CO(NH$_2$)$_2$) lactic acid, sodium hydroxide, ammonia solution (25% w/w), hydrochloric acid, glycerol, and methanol were purchased from Merck company (Darmstadt, Germany). Ethylene glycol bisthioglycolate (EGBTG) was purchased from Sigma chemical (St Louis, MO, USA) and deionized water was used through the experiments. All chemicals and reagents were of analytical grade and used as supplied.

Instrumentation

Determination of metal ions was carried out by a Varian Vista-Pro ICP-OES (Springvale, Australia). The instrument was operated at applied power of 1.2 kW and plasma and argon flow rates were 13.5 and 0.75 L min^{-1}, respectively. Transmission electrom microscope (TEM) images of the adsorbent were obtained using a Philips CM200 transmission

electron microscope (Amsterdam, Netherlands). The X-ray diffraction (XRD) analysis of modified nanoparticles was performed by an ItalStructures APD 2000 X-ray diffractometer (Riva Del Garda, Italy) using Cu K$_\alpha$ radiation source λ 1.540598 A° with reflection scan mode. A Metrohm 827 pH/mV meter (Herisau, Switzerland) with a combined glass electrode was used for pH measurements.

Preparation of artificial saliva

The artificial saliva solution was prepared as described by Fusayama et al. [20, 21] in final volume of 1000 mL and its composition was as NaCl (0.40 mg mL^{-1}), CaCl$_2$.2H$_2$O (0.79 mg mL^{-1}), KCl (0.40 mg mL^{-1}), NaH$_2$PO$_4$.2H$_2$O (0.78 mg mL^{-1}), Na$_2$S.9H$_2$O (0.005 mg mL^{-1}), CO (NH$_2$)$_2$ (1.00 mg mL^{-1}). The pH of the prepared artificial saliva was 5.5. In recommended procedure, 5 mL of the solution was diluted to 200 mL and lactic acid and ammonia solutions were used to adjust saliva pH in the recommended MSPE step.

Preparation of magnetic adsorbent

The Au@Mn-Fe$_3$O$_4$ NPs were prepared based on the reported method in the literature [22]. Briefly, ferrous chloride tetrahydrate (2.15 g) and ferric chloride hexahydrate (5.84 g) were dissolved in 100 mL deionized water under nitrogen atmosphere with vigorous stirring at 90 °C. Then, 30 mL of ammonia solution was added and the mixture was stirred for 15 min at the same condition. The prepared Fe$_3$O$_4$ NPs were cooled to the room temperature. The precipitate was isolated from the solution by applying an external magnet and the supernatant was removed. Then, the precipitate was washed with deionized water (3 ×250 mL) and 0.01 mol L^{-1} sodium chloride solution (2 ×100 mL). The later can accelerate magnetic separation particularly in alkaline solutions [23, 24]. Then, the suspension was transferred to a 250 mL round bottom flask and allowed to settle. The supernatant was removed and an aqueous solution of MnSO$_4$ (10 mL, 0.1 M) was added to the mixture and stirred for 30 min, followed by the addition of KMnO$_4$ (10 mL, 0.1 M) as oxidant. The mixture was then stirred for 1 h and the precipitate (Mn-Fe$_3$O$_4$ NPs) was washed with 200 mL of deionized water by decantation using supermagnet. After that, 5 mL of 5 mM HAuCl$_4$ was added to the mixture and allowed to boil with constant stirring under reflux. The reaction was followed by addition of 10 mL of 25 mM trisodium citrate under continuous stirring. The mixture was

remained at the same condition for 15 min and then rapidly cooled to the room temperature using an ice bath. In the next step, the supernatant was removed and an aqueous solution of EGBTB (1.0% v/v, 150 mL) was added followed by glycerol (60 mL). The mixture was stirred at 90 °C for 2 h. After cooling down slowly to the room temperature, the precipitate was easily isolated from the supernatant using a supermagnet and the suspension was washed sequentially with deionized water (250 mL, three times), methanol (200 mL, twice), and deionized water (250 mL, once) before it was dried in vacuum oven at 50 °C for 24 h.

Recommended MSPE procedure

A portion of artificial saliva solution containing 5 µg of target metal ion was transferred to a 250 mL beaker. The pH was adjusted by lactic acid or/and ammonia solution and diluted to the final volume of 200 mL. Then, 100 mg of Au@Mn-Fe$_3$O$_4$ NPs adsorbent was added and the solution was stirred for 5 min. The mixture was placed on a supermagnet and the supernatant was completely decanted. The magnet was removed; 2 mL of 0.1 M HCl solution was added and the mixture was stirred for 2 min. The solution was placed on a supermagnet again to hold the adsorbent in the bottom of the flask and the supernatant was transferred to a test tube for the subsequent ICP-OES analysis.

Method validation

Linearity was evaluated by analyzing different concentrations of metal ions in the range of 5-250 ng mL^{-1} and calibration curves (five points) were constructed. Each point is the mean value of three-replicate analyses. The slope, intercept and correlation coefficient were calculated using the least square regression method. The limit of detection (LOD) and limit of quantitation (LOQ) were calculated based on the standard deviation of blank (S_b) and slope of calibration curve (m) determined according to the following equations:

$$LOD = 3.3S_b/m \tag{1}$$

$$LOQ = 10S_b/m \tag{2}$$

Precision of the proposed method was determined by five-replicate analyses of artificial saliva spiked with each analyte at 50 ng mL^{-1}. Preconcentration factor (PF) of the method was calculated as PF = V$_s$/V$_e$× R%, where V$_s$ is the sample volume, V$_e$ is elution volume, and R% is percentage recovery of each analyte.

RESULT AND DISCUSSION

Soft coordination sites like *SH* groups are known to have a great tendency to soft metal ions, such as Ag^+ and Pb^{2+}. This work shows that combination of high adsorbent capacity of Mn-Fe_3O_4 NPs and modification with an *S* donor ligand significantly increase the quantitative retention of these ions as well as lowering their detection limits.

Characterization of adsorbent

Brunauer–Emmett–Teller (BET) analysis of Fe_3O_4 NPs before and after doping with Mn shows an increased surface area from 47 m^2 g^{-1} to 73 m^2 g^{-1}, respectively, explaining the increase in analyte affinity of the adsorbent. Fig. 1 shows TEM image of the prepared adsorbent to determine its size and morphology. As can be seen , the prepared adsorbent has uniform size distribution with average diameter of 40±5 nm while most of the particles are quasi spherical in shape. The XRD patterns of the standard Fe_3O_4 particles and the prepared adsorbent before and after doping with Mn and modifying with Au NPs are presented in Fig. 2. This figure shows the Brag reflections indexed to the inverse cubic spinel phase of iron oxide for both samples which match well with standard magnetite obtained by JCPDS [25, 26]. There was no indication of additional phases and each sample contains identical peaks regardless of Mn content. There is no evidence of peaks

corresponding to MnO_2 or Mn_3O_4, confirming that Mn is incorporated into the ferrite structure rather than precipitating as oxides on the surface of Fe_3O_4 NPs. Further, peak positions of Mn-Fe_3O_4 NPs were shifted to lower peak position which also suggesting that the Fe^{3+} ions are effectively substituted by larger radius Mn^{2+} ions [27]. For Au@Mn-Fe_3O_4 NPs two additional peaks corresponding to 111 and 200 plans of
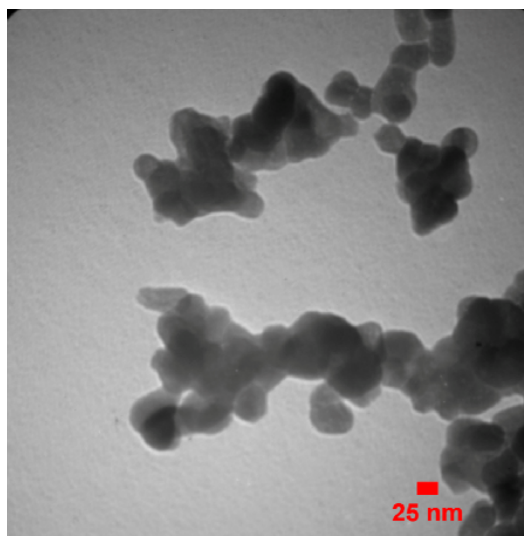

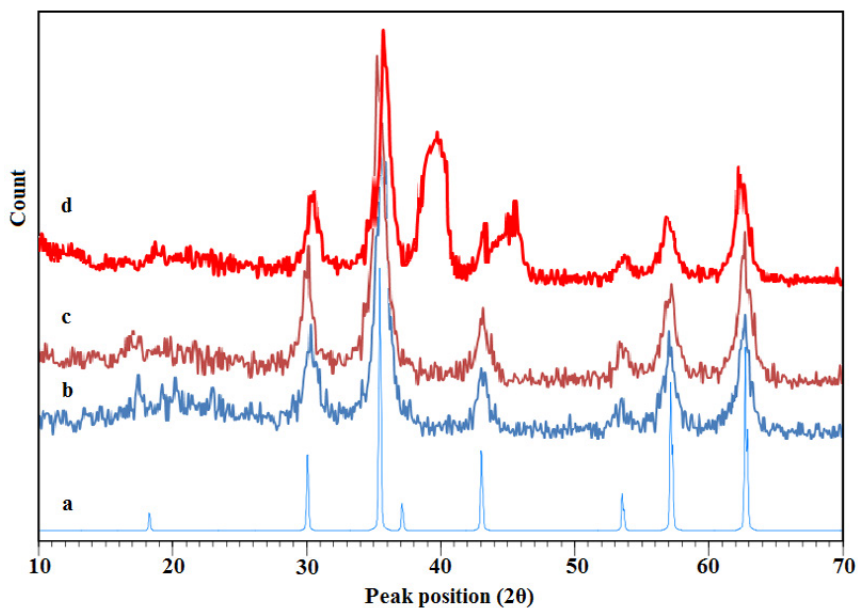Fig. 1: TEM image of the prepared adsorbent with mean diameter of 40 nm.

Fig. 2: XRD patterns of (a) standard of Fe_3O_4, (b) prepared Fe_3O_4 NPs, (c) Mn-Fe_3O_4 NPs and (d) Au@ Mn-Fe_3O_4 NPs.

Au NPs were observed at peak positions about 37.7 and 44.5 [28]. The saturation magnetization curves of Fe$_3$O$_4$ NPs, Mn-Fe$_3$O$_4$ NPs and Au@Mn-Fe$_3$O$_4$ NPs are shown in Fig. 3. As can be seen, Fe$_3$O$_4$ NPs has a saturation magnetization about 43 emu g^{-1}. The saturation magnetization of Au@Mn-Fe$_3$O$_4$ NP adsorbents was decreased due to the thickening of the shells, however its magnetization still shows 24 emu g^{-1}, which is more than 56% of Fe$_3$O$_4$ NPs' magnetization. This appears to be due to the oxidative doping process removing Fe^{3+} ions from the lattice and incomplete replacement with the larger radius Mn ions, resulting in a more disordered structure and subsequent decrease in magnetic strength which confirms the results obtained by XRD analysis. However, the prepared adsorbent has enough magnetic responsiveness for applications which need magnetic manipulation.

Effect of pH

Optimum pH value which is required for quantitative extraction of the target ions was evaluated in the range of 2.0-7.0 for 5 μg mL^{-1} of each metal ion in artificial saliva solution. Higher pH was not considered due to probability of hydroxide precipitation, and the results were shown in Fig. 4. As can be seen, quantitative absorption was obtained in different pH values for each metal ion. Quantitative efficiency was obtained in pH 5.0 for Ag$^+$, and it remains constant in higher pHs. Maximum adsorption of Pb^{2+} was obtained in the range of 4.5-7.0. Thus, optimum pH for simultaneous extraction of Ag+ and Pb^{2+} ions in artificial saliva ranged from 5.0-7.0. Considering the fact that normal pH of artificial saliva is 5.5 and -SH groups of EGBTG modifier can be oxidized in low pH, the value of 5.5 was selected for all our subsequent experiments including natural saliva samples.
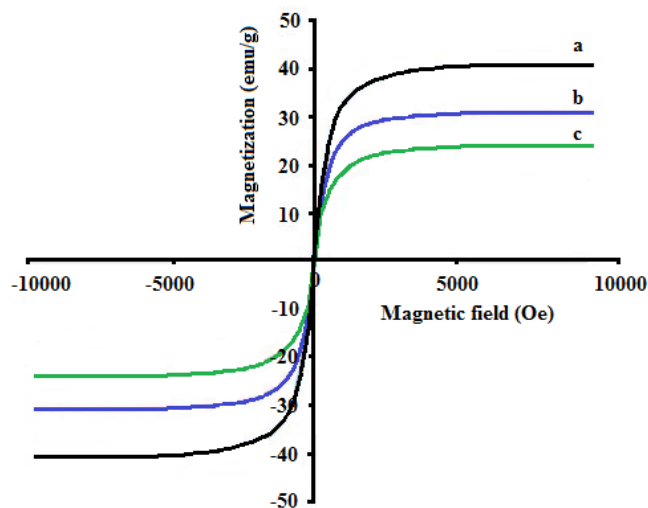

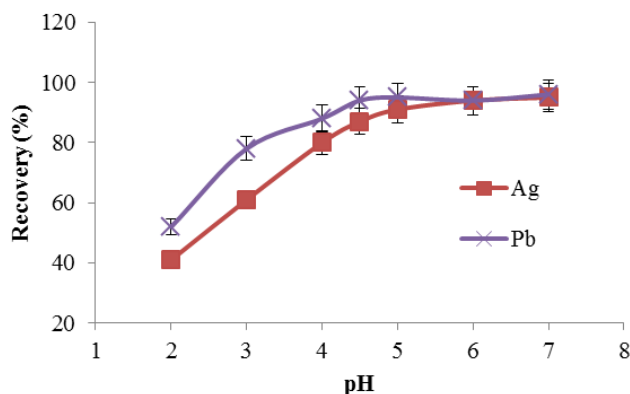Fig. 3: VSM analysis of Fe$_3$O$_4$ NPs, Mn-Fe$_3$O$_4$ NPs and Au@Mn-Fe$_3$O$_4$ adsorbent.

Fig. 4: Effect of pH on the recovery of targeted metal ions. Sample volume; 200 mL, adsorbent amount; 100 mg, extraction time; 5 min, desorption time; 2 min, and eluent solvent; 0.1 M HCl.

Effect of Eluent

It was revealed from the pH studies that the adsorbent shows low tendency to target metal ions in high acidic media. Thus, the effect of desorbing solvent on the recovery of target ions was investigated by choosing some strong acids such as 0.1 M HNO_3, 0.1 M HCl, and 0.1 M H_2SO_4 and some organic solvents such as ethanol and acetonitrile. The experimental results demonstrated that a solution of 0.1 M HCl has higher elution efficiency with respect to the other eluent. Therefore, it was selected for all subsequent experiments. On the other hand, eluent volume is a critical parameter affecting on the preconcentration factor and extraction kinetics. Thus, the effect of eluent volume for quantitative elution of target ions was investigated by changing it in the range of 0.5-10 mL. Based on the results, minimum volume for quantitative elution of the retained analytes was 2.0 mL. Thus, this value was selected for the following experiments.

Effect of the sample volume

Large sample volume and low eluent volume need to achieve higher preconcentration factor. The effect of sample volume on the extraction efficiency of targeted metal ions was studied by applying recommended MSPE procedure to different sample volumes of artificial saliva (25-250 mL) containing 5.0 μg of each metal ion. The results indicated that quantitative recovery of Ag^+ and Pb^{2+} ions were achieved with maximum sample solution volume of 200 mL. Decreasing in the extraction recoveries can be explained by considering mass-balance equation. Theoretical recovery (R) can be calculated using R = 1/1+(β/$K_{s,w}$) [29], where, β is phase ratio

(sample to adsorbent volume ratio) and $K_{s,w}$ is the partition coefficient. From the equation, when the sample volume is too large, β increases and causes decreasing in recovery. Therefore, 200 mL was selected for sample volume in next experiments.

Adsorbent amount and adsorbent capacity

NPs have significantly higher surface area and short diffusion route compared to the traditional adsorbents which can result in high extraction efficiency and fast extraction dynamics. Hence, satisfactory results can be obtained with lower adsorbent amount. To obtain minimum adsorbent amount needed for quantitative extraction of target analytes, the amount of Au@Mn-Fe_3O_4 NPs was varied in the range of 10-200 mg. The results revealed that quantitative recoveries were obtained by using 100 mg of the adsorbent and this value was selected for the next experiments.

On the other hand, one of the most advantages of using modified MNPs is higher adsorption capacity which is due to their very high surface area. The capacity of an adsorbent determines how much adsorbent is requiring for quantitative extraction of certain amount of metal ions from the solution. In order to determine the adsorption capacity, 100 mg of Au@Mn-Fe_3O_4 NPs and 200 mL of various concentrations of metal ions were examined and the concentrations were increased until saturation occurred at equilibration time of 15 min. The maximum adsorption capacity was found to be 14.9 and 19.1 mg g^{-1} for Ag^+ and Pb^{2+} ions, respectively. The results indicated that adsorption capacity of the prepared adsorbent was much higher than the required amount for determination of these ions in biological samples such as saliva.
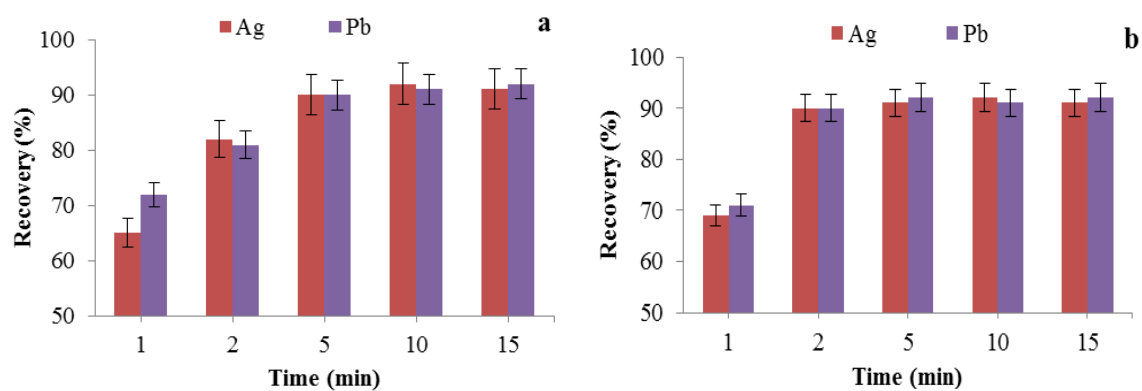

Fig. 5: Effect of extraction and desorption times on the recovery of targeted metal ions. Sample volume; 200 mL, adsorbent amount; 100 mg, eluent solvent; 0.1 M HCl and pH; 5.5.

Effect of extraction and desorption times

Efficient dispersion of the adsorbent can considerably increase its contact surface with the analyte leading to a higher adsorption efficiency and faster extraction time. Nanomaterials have higher surface area with respect to the traditional adsorbents and can be rapidly collected from the sample solution by using a magnetically assisted separation procedure. Hence, fast extraction and desorption times can be achieved with these materials. Fig. 5 shows the recovery values of metal ions as a function of extraction and desorption times. Based on the results, 5 min is sufficient for achieving an appropriate adsorption and 2 min was enough for a quantitative desorption, which are significantly shorter times compared to those in traditional column-passing SPE.

Adsorbent reusability

Reusability is one of the key parameters for evaluation of an adsorbent performance. In order to determine regeneration capability of Au@Mn-Fe₃O₄ NPs, the adsorbent which previously used in the general MSPE procedure was washed with desorbing solvent (10 mL, twice) and deionized water (10 mL, five times), then dried in oven at 50 ºC and reused. Experimental results revealed that the adsorbent can be reused at least up to ten times (RSD <3.9%) without sacrifice the analytical performance which makes it a suitable alternative to conventional SPE adsorbent.

Analytical performance

Under the optimum conditions, the target ions were quantitatively extracted from spiked artificial saliva by applying the recommended MSPE procedure and the resulted analytical characteristics are summarized in Table 1. Linearity of the proposed method was obtained in the range

of 5-250 ng mL⁻¹ with correlation coefficients (R^2) of 0.991. As can be seen from the table, LODs were obtained 0.09 and 0.11 ng mL⁻¹ and the LOQs were obtained 0.27 and 0.32 ng mL⁻¹ for Ag^+ and Pb^{2+} ions, respectively. Relative standard deviation (RSD %) of the method for determination of the analytes was calculated 3.5 and 3.7 %, respectively. The enrichment factor for each metal ion was found to be 94 and 95 for Ag^+ and Pb^{2+}, respectively.

Application of the method

To investigate the accuracy and applicability of the optimized method in real biological samples, the concentration of Ag^+ and Pb^{2+} ions was determined in saliva samples obtained from 9 volunteers (3 males and 6 females) with average of 19 years old and in the range of 15-29 years old that had no amalgam fillings. The informed consent was obtained from all volunteers at the beginning of the study and before fillings. Then, in 24, 72 and 96 hours after that one of their posterior decayed teeth was filled with amalgam, 2 mL of their saliva were collected by spitting in a special tube and kept at -18 ºC in a freezer immediately. Once all the samples were collected, they were transferred to the laboratory for analysis. Before testing, the samples were transferred into the three 200 mL flasks, put in three groups and labeled to saliva *I, II* and *III*. If needed, pH was adjusted and then diluted to the volume. Samples labeled saliva *I* were analyzed without any treatment and samples labeled saliva *II* and *III* were spiked with 10 and 100 ng mL⁻¹ of each metal ion, respectively, for recovery tests, and then, were analyzed with the recommended MSPE procedure. The short-term release of the targeted metal ions was summarized in Table 2. As can be seen, the recoveries of target metal ions were in the range of 94 to 97% and the method has good merit for determination of these metal ions in biological high saline saliva.

Table 1: Results for determination of analytical parameters for determination of Ag^+ and Pb^{2+} ions using the recommended MSPE procedure.

Analyte	Calibration equation	R^2	LOD	LOQ	RSD (%)	Recovery (%)
Ag^+	Y=3012C-874	0.993	0.09	0.27	3.5	94.4
Pb^{2+}	Y=2122C-712	0.991	0.11	0.32	3.7	95.3

Table 2: Results for the determination of targeted of Ag^+ and Pb^{2+} ions in saliva samples (n=5).

Sample	Found (ng mL⁻¹), RSD%		Recovery (%)	
	Ag^+	Pb^{2+}	Ag^+	Pb^{2+}
Saliva I	67.6, 3.1	11.4,3.9	-	-
Saliva II	72.9, 2.7	20.1, 3.3	94	94
Saliva III	160.9, 2.8	108.0, 2.9	96	97

CONCLUSION

An efficient modified adsorbent as Au NPs@Mn-Fe$_3$O$_4$ NPs was synthesized and successfully used for extraction and preconcentration of Ag$^+$ and Pb^{2+} ions released to saliva of volunteers which were exposed to amalgam filling of their posterior decayed teeth. The method combines high adsorption capacity of the NPs and magnetic separation, and provides a fast, convenient, effective, and sensitive method for preconcentration and separation of these metal ions in high saline samples. In addition, reusability studies demonstrated the potential of the prepared adsorbent for recyclable usage compared to the conventional column-based SPE. The method is simple and inexpensive, and eliminates the need to use any auxiliary facilities to immobilize the adsorbent.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

REFERENCES

1. Ahmed HS, Zaidan TF, Yakub A. Trace elements and oxidative stress markers in saliva of subjects with amalgam fillings. Journal of Baghdad College of Dentistry. 2012;24(3):63-6.
2. Campus G, Garcia-Godoy F, Gaspa L, Panzanelli A, Piu PC, Micera G, et al. Dependence of kinetic variables in the short-term release of Hg2+, Cu2+ and Zn2+ ions into synthetic saliva from an high-copper dental amalgam. Journal of Materials Science: Materials in Medicine. 2007;18(8):1521-7.
3. Al-Salehi SK, Hatton PV, McLeod CW, Cox AG. The effect of hydrogen peroxide concentration on metal ion release from dental amalgam. Journal of Dentistry. 2007;35(2):172-6.
4. Mutlu I, Oktay E. Characterization of 17-4 PH stainless steel foam for biomedical applications in simulated body fluid and artificial saliva environments. Materials Science and Engineering: C. 2013;33(3):1125-31.
5. Mareci D, Chelariu R, Dan I, Gordin D-M, Gloriant T. Corrosion behaviour of β-Ti20Mo alloy in artificial saliva. Journal of Materials Science: Materials in Medicine. 2010;21(11):2907-13.
6. Sanna G, Pilo M, Piu P, Spano N, Tapparo A, Campus G, et al. Study of the short-term release of the ionic fraction of heavy metals from dental amalgam into synthetic saliva, using anodic stripping voltammetry with microelectrodes. Talanta. 2002;58(5):979-85.
7. Fields C, Mallee P, Muzard J, Lee GU. Isolation of Bowman-Birk-Inhibitor from soybean extracts using novel peptide probes and high gradient magnetic separation. Food Chemistry. 2012;134(4):1831-8.
8. Yang X, Zhang X, Ma Y, Huang Y, Wang Y, Chen Y. Superparamagnetic graphene oxide–Fe3O4 nanoparticles hybrid for controlled targeted drug carriers. Journal of Materials Chemistry. 2009;19(18):2710.
9. Huh Y-M, Jun Y-w, Song H-T, Kim S, Choi J-s, Lee J-H, et al. In Vivo Magnetic Resonance Detection of Cancer by Using Multifunctional Magnetic Nanocrystals. Journal of the American Chemical Society. 2005;127(35):12387-91.
10. Shabanian M, Khoobi M, Hemati F, Khonakdar HA, ebrahimi SeS, Wagenknecht U, et al. New PLA/PEI-functionalized Fe3O4 nanocomposite: Preparation and characterization. Journal of Industrial and Engineering Chemistry. 2015;24:211-8.
11. Aghahosseini H, Ramazani A, Ślepokura K, Lis T. The first protection-free synthesis of magnetic bifunctional l -proline as a highly active and versatile artificial enzyme: Synthesis of imidazole derivatives. Journal of Colloid and Interface Science. 2018;511:222-32.
12. Khoobi M, Delshad TM, Vosooghi M, Alipour M, Hamadi H, Alipour E, et al. Polyethyleneimine-modified superparamagnetic Fe3O4 nanoparticles: An efficient, reusable and water tolerance nanocatalyst. Journal of Magnetism and Magnetic Materials. 2015;375:217-26.
13. Motevalizadeh SF, Khoobi M, Sadighi A, Khalilvand-Sedagheh M, Pazhouhandeh M, Ramazani A, et al. Lipase immobilization onto polyethylenimine coated magnetic nanoparticles assisted by divalent metal chelated ions. Journal of Molecular Catalysis B: Enzymatic. 2015;120:75-83.
14. Tarasi R, Khoobi M, Niknejad H, Ramazani A, Maʼmani L, Bahadorikhalili S, et al. β -cyclodextrin functionalized poly (5-amidoisophthalicacid) grafted Fe 3 O 4 magnetic nanoparticles: A novel biocompatible nanocomposite for targeted docetaxel delivery. Journal of Magnetism and Magnetic Materials. 2016;417:451-9.
15. Tarasi R, Ramazani A, Ghorbanloo M, Khoobi M, Aghahosseini H, Joo SW, et al. Preparation and Characterization of MCM-41@PEI·Mn as a New Organic-Inorganic Hybrid Nanomaterial and Study of its Catalytic Role in the Oxidation of Cyclohexene, Ethyl Benzene, and Toluene in the Presence of H2O2as an Oxidant. Bulletin of the Korean Chemical Society. 2016;37(4):529-37.
16. Yang B, Gong Q, Zhao L, Sun H, Ren N, Qin J, et al. Preconcentration and determination of lead and cadmium in water samples with a MnO2 coated carbon nanotubes by using ETAAS. Desalination. 2011;278(1-3):65-9.
17. Han R, Zou W, Li H, Li Y, Shi J. Copper(II) and lead(II) removal from aqueous solution in fixed-bed columns by manganese oxide coated zeolite. Journal of Hazardous Materials. 2006;137(2):934-42.
18. Liu X, Wu N, Cui C, Bi N, Sun Y. One pot synthesis of Fe3O4/MnO2 core–shell structured nanocomposites and their application as microwave absorbers. RSC Advances. 2015;5(31):24016-22.
19. O'Hara MJ, Carter JC, Warner CL, Warner MG, Addleman RS. Magnetic iron oxide and manganese-doped iron oxide nanoparticles for the collection of alpha-emitting radionuclides from aqueous solutions. RSC Advances. 2016;6(107):105239-51.
20. Shimazu K, Karibe H, Ogata K. Effect of artificial saliva contamination on adhesion of dental restorative materials. Dental Materials Journal. 2014;33(4):545-50.
21. Baumann T, Kozik J, Lussi A, Carvalho T. Erosion protection conferred by whole human saliva, dialysed saliva, and artificial saliva. Scientific reports. 2016;6:34760.
22. Mashhadizadeh MH, Amoli-Diva M, Pourghazi K. Magnetic nanoparticles solid phase extraction for determination of ochratoxin A in cereals using high-performance liquid chromatography with fluorescence detection. Journal of Chromatography A. 2013;1320:17-26.

23. Chang Y-C, Chen D-H. Preparation and adsorption properties of monodisperse chitosan-bound Fe3O4 magnetic nanoparticles for removal of Cu(II) ions. Journal of Colloid and Interface Science. 2005;283(2):446-51.

24. Liao M-H, Chen D-H. Preparation and characterization of a novel magnetic nano-adsorbent. Journal of Materials Chemistry. 2002;12(12):3654-9.

25. Cheng Q, Qu F, Li NB, Luo HQ. Mixed hemimicelles solid-phase extraction of chlorophenols in environmental water samples with 1-hexadecyl-3-methylimidazolium bromide-coated Fe3O4 magnetic nanoparticles with high-performance liquid chromatographic analysis. Analytica Chimica Acta. 2012;715:113-9.

26. Mashhadizadeh MH, Amoli-Diva M. Atomic absorption spectrometric determination of Al3+ and Cr3+ after preconcentration and separation on 3-mercaptopropionic acid modified silica coated-Fe3O4 nanoparticles. Journal of Analytical Atomic Spectrometry. 2013;28(2):251.

27. Suresh R, Giribabu K, Manigandan R, Vijayalakshmi L, Stephen A, Narayanan V. Electrochemical sensing property of Mn doped Fe[sub 3]O[sub 4] nanoparticles. AIP; 2013.

28. Xing Y, Jin Y-Y, Si J-C, Peng M-L, Wang X-F, Chen C, et al. Controllable synthesis and characterization of Fe3O4/Au composite nanoparticles. Journal of Magnetism and Magnetic Materials. 2015;380:150-6.

29. Cheng G, He M, Peng H, Hu B. Dithizone modified magnetic nanoparticles for fast and selective solid phase extraction of trace elements in environmental and biological samples prior to their determination by ICP-OES. Talanta. 2012;88:507-15.