CHAPTER IV

RESULTS AND DISCUSSION

The synthesis and characterization of materials are first described. The extraction properties of materials are next discussed in both batch and column methods. In batch method, various parameters such as type of metal, pH of sample solution and the presence of divers ions in metal solution are examined, while the amounts of metal, the concentration and the volume of eluent are discussed in the column method. Finally, the application of HPMSP doped mesoporous silica to the extraction of metal in food sample is shown.

4.1. Synthesis of materials

The synthesis of non-doped and HPMSP doped mesoporous silica were carried out according to the procedure reported by Boos *et al.* [18]. The molar composition used for the synthesis was 1 TEOS : 140 H_2O (0.1 M NaOH) : 13 MeOH : 0.18 CTAB. For HPMSP doped mesoporous silica, 0.06 mole of HPMSP per 1 mole of TEOS were also used for the synthesis. It was found that all HPMSP were successfully incorporated in the silica without any release since no absorption spectra of HPMSP were observed in the UV spectra of the supernatant and washing solutions. This observation was in accordance with those reported by Boos *et al.* [18].

4.2. Characterization of materials

4.2.1. Determination of organic matter contents

The organic matter contents in both non-doped and HPMSP doped mesoporous silica determined by calcination (namely measured values) and those calculated from starting materials (namely calculated values) from six synthesis experiments were shown in Table 4.1. It is obviously seen that the measured values of organic matters in both non-doped and HPMSP doped mesoporous silica were slightly higher than the calculated values. The minor difference may be explained by the amounts of physisorbed water and chemisorbed water, which were remained in the silica framework before the beginning of calcination experiment. In fact, the removal of physisorbed water and chemisorbed water was achieved at 170°C and 400°C, respectively [49].

Table 4.1 Amount of organic matters in non-doped and HPMSP doped mesoporous silica.

Type of silica	Organic matter contents (%)		$\frac{\text{Measured values}}{\text{Calculated values}}$ x 100 (%)
	Measured values	Calculated values*	
Non-doped	47.94 ± 0.90	45.93 ± 0.40	104.33 ± 2.33
HPMSP doped	58.85 ± 0.69	56.54 ± 0.17	104.09 ± 1.15

* The calculated values of organic matters were determined and shown in Appendix C.

4.2.2 Crystallinity of materials

The crystallinity of materials was investigated for both as-synthesized and corresponding calcined mesoporous silica using X-ray diffraction technique. The diffractograms of materials studied were shown in Figure 4.1.

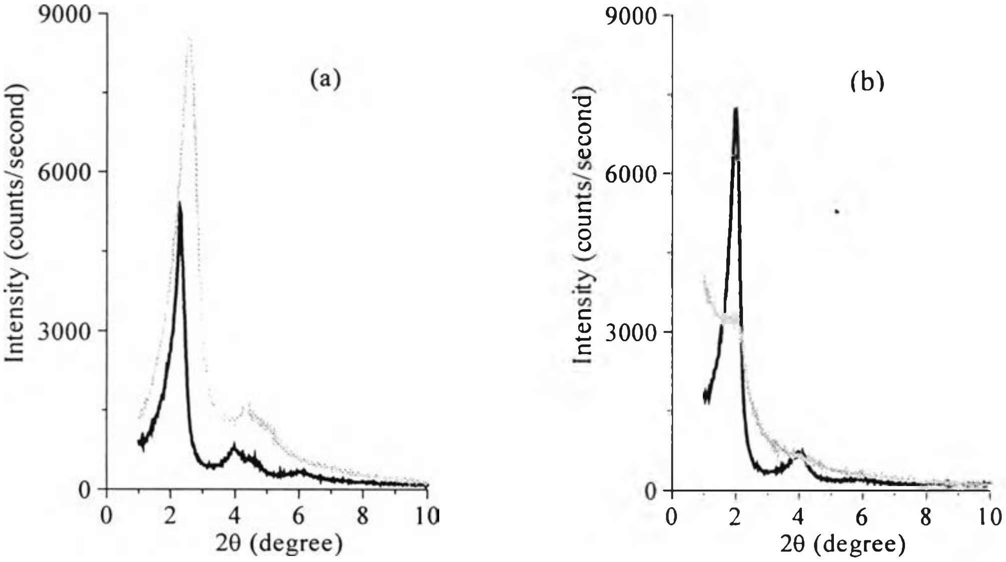

Figure 4.1 The XRD patterns of (a) non-doped mesoporous silica and (b) HPMSP doped mesoporous silica (black: as-synthesized silica, gray: calcined silica).

From Figure 4.1, the XRD patterns of the as-synthesized non-doped and HPMSP doped mesoporous silica exhibited a very strong (100) reflection peak at low-angle degree. The presence of this reflection peak indicated the periodic structure of both materials. This observation was in accordance with the results reported by Boos *et al.* [18]. After calcination, a (100) reflection peak was still observed in case of non-doped mesoporous silica while the peak was broadened in the case of HPMSP doped mesoporous silica. These results indicated the thermal resistance of non-doped mesoporous silica structure and the degradation of crystallinity of HPMSP doped mesoporous silica after treatment at high temperature. In addition, the d-spacing values of both as-synthesized and calcined materials were compared and tabulated in Table 4.2. As seen from this table, the d-spacing value at 2θ around 2^{o} of the as-synthesized HPMSP doped silica was slightly higher than that of non-doped silica. This result may be caused by the HPMSP molecules incorporated in silica that expanded the distance between each lamellar layer of silica.

Table 4.2 The d-spacing values of as-synthesized and calcined mesoporous silica.

Types of silica	As-synthesized silica		Calcined silica	
	2θ (degree)	d-spacing (Å)	2θ (degree)	d-spacing (Å)
Non-doped	2.30 3.98	38.38 22.18	2.58 4.44	34.22 19.88
HPMSP doped	1.98 4.10	44.56 21.53	-	-

4.2.3. Mesoporosity, surface area and pore size

Nitrogen sorption measurement is another method used for the determination of mesoporosity, surface area and pore size of materials. The specific surface area of both non-doped and HPMSP doped mesoporous silica was calculated using BET method. Their total pore volume was obtained from the desorption volume at P/P_0 = 0.98 and the pore size distribution was calculated from BJH equation. The obtained N_2 sorption isotherms were displayed in Figure 4.2.

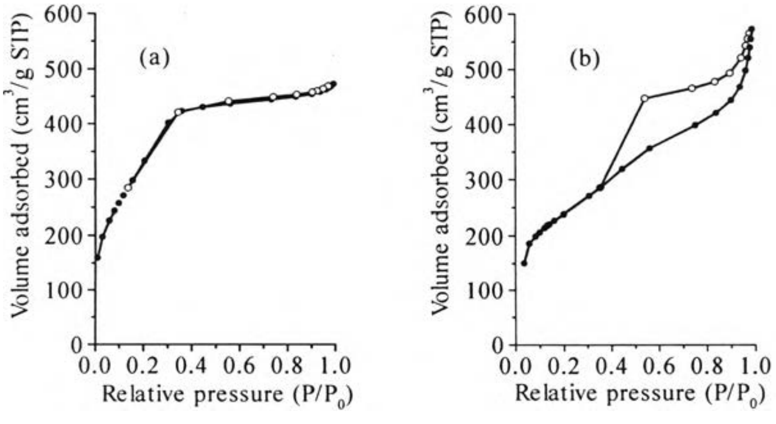

Figure 4.2 N_2 adsorption-desorption isotherms of (a) non-doped mesoporous silica and (b) HPMSP doped mesoporous silica (–•–: adsorption, –o–: desorption).

As seen from Figure 4.2, both materials showed irreversible type IV adsorption isotherms with a H4 hysteresis loop as defined in the IUPAC [23]. A linear increase of adsorbed volume at low pressure followed by a steep increase in nitrogen uptake at a relative low pressure was observed. These phenomena were typically a characteristic of the capillary condensation inside the mesopores. This result was in accordance with the previous work reported by Boos and co-workers [18]. For the BJH pore size distribution of materials, the results were illustrated in Figure 4.3. It was found that both materials had narrow pore size distribution. Furthermore, the average pore size of non-doped mesoporous silica was 27.3 Å whereas they were found to be 38.5 Å for HPMSP doped mesoporous silica. The higher values in pore size of HPMSP modified materials may be originated by the presence of HPMSP molecules in the silica as described previously. In addition, the textural properties obtained from the N_2 sorption measurement were also summarized in Table 4.3. It is clear that the incorporation of HPMSP molecules into the silica made the pore volume and pore diameter larger. Also, these results were consistent with those previously reported by Boos *et al.* [18].

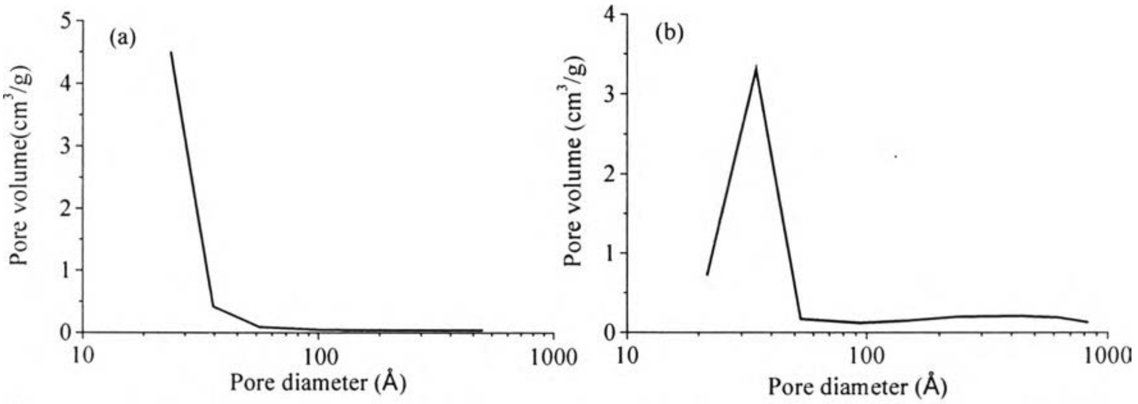

Figure 4.3 BJH pore size distribution of (a) non-doped mesoporous silica and (b) HPMSP doped mesoporous silica.

Table 4.3 The textural properties of calcined non-doped and HPMSP doped mesoporous silica.

Type of silica	Surface area, S (m^2/g)	Pore volume, Vp (cm^3/g)	APD (Å)
Non-doped	1059	0.724	27.3
HPMSP doped	867	0.835	38.5

S, BET surface area obtained from N_2 sorption; V_p, total pore volume obtained from single-point volume at $P/P_0 = 0.98$; APD, average pore diameter calculated from 4Vp/S.

4.3. Extraction properties of materials

The metal extraction efficiency of mesoporous silica could be influenced by various parameters such as types of metal, pH of metal solution, the presence of diverse ions in metal solution, etc. Thus, to obtain the optimum extraction condition, various experimental parameters were performed in both batch and column methods. The results of each extraction process were described below.

4.3.1. Batch method

4.3.1.1. Reproducibility of materials for the extraction of metal

According to Boos and co-worker [18], the optimum condition used for the extraction of Cu(II) by HPMSP doped mesoporous silica was at pH 2 with the presence of 0.1 M $NaNO_3$. Therefore, the same condition was adopted in this study with the initial concentration of Cu(II) equal to 200 ppm. The extraction experiment was managed using HPMSP doped mesoporous silica from different lots. Each lot comprised eight syntheses. The reproducibility results were displayed in Table 4.4. As seen, the average Cu(II) extraction capacity was 0.2035 ± 0.0072 mol/kg with 3.51% of relative standard deviation. This small RSD value indicated the excellent reproducibility of HPMSP doped mesoporous silica.

Since the Cu(II) ions could not be extracted by the non-doped mesoporous silica, the target metal was thus replaced by Zn(II) ions in order to determine the reproducibility of this sorbent. The $Zn(OAc)_2$ was used as metal source and the extraction medium was at pH 3 with the presence of 0.1 M $NaNO_3$. The initial concentration of Zn(II) was 200 ppm. The experiments were performed on both non-doped and HPMSP doped mesoporous silica. The obtained results were tabulated in Table 4.5. From that table, both materials had good reproducibilities of Zn(II) extraction since the relative standard deviation was quite low. It is also noteworthy that the Zn(II) extractability of the HPMSP modified silica was higher than that of non-doped material. This event could be explained by the presence of HPMSP in the sorbent. Indeed, these functionalized molecules can form stable complexes with the Zn(II) ions [50].

Table 4.4 The reproducibility results of Cu(II) extraction by HPMSP doped mesoporous silica from different lots.

Lot number	Cu(II) extracted (mol/kg)
1	0.1919
2	0.1993
3	0.2027
4	0.1953
5	0.2029
6	0.2137
7	0.2063
8	0.2082
9	0.2108
Average	0.2035
SD	0.0072
%RSD	3.51

Table 4.5 The reproducibility results of Zn(II) extraction by non-doped or HPMSP doped mesoporous silica from different lots.

Lot number	Zn(II) extracted (mol/kg)	
	Non-doped silica	HPMSP doped silica
1	0.1948	0.2768
2	0.2080	0.2821
3	0.2266	0.2813
4	0.2021	0.2850
5	0.2029	0.3009
6	0.2005	0.2940
7	0.2098	0.3087
8	0.2290	0.2959
9	-	0.3138
10	-	0.3211
Average	0.2092	0.2960
SD	0.0123	0.0150
%RSD	5.91	5.08

4.3.1.2. Effect of pH

HPMSP is an acidic chelating agent. The reaction between HPMSP and metal ions can be expressed as follows.

$$M^{n+} + nHPMSP \rightleftharpoons M(PMSP)_n + nH^+$$

where $M(PMSP)_n$ is the metal-chelating agent complex. The above reaction is a reversible reaction, wherein a cation and hydrogen ions are exchanged. Thus, the degree of extraction of metal ions depends on the pH of the metal solution and that pH could affect the stability of the complex.

In this work, the influence of pH on the metal extraction efficiency of non-doped and HPMSP doped mesoporous silica was investigated. The target metals included Cd(II), Pb(II) and Zn(II). The desired pH of metal solution was adjusted using 1.0 M HNO_3. The results of each metal were described below.

i. Extraction of Cd(II)

The influence of pH on the Cd(II) extraction was studied in the pH range of 2 to 6. The amounts of Cd(II) extracted were plotted against initial pH of metal solution as displayed in Figure 4.4.

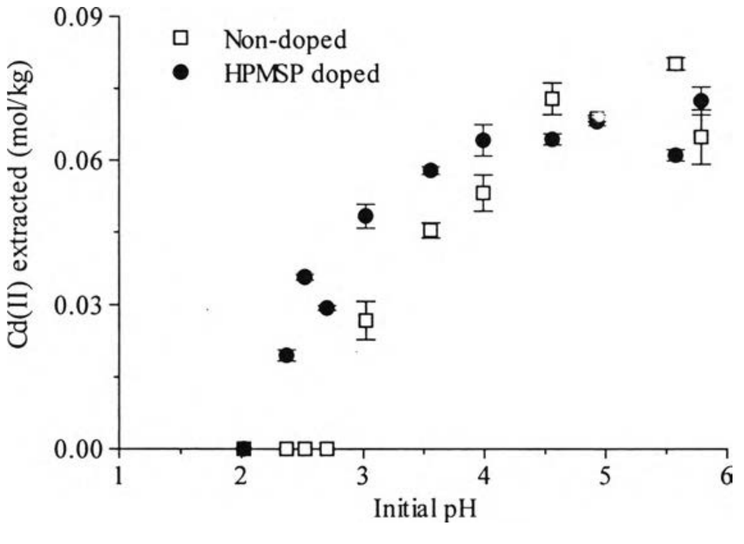

Figure 4.4 Effect of pH on the Cd(II) extractability of non-doped and HPMSP doped mesoporous silica.

From Figure 4.4, the Cd(II) extractability of both materials seemed to be dependent on the initial pH of metal solution. The non-doped mesoporous silica began to extract Cd(II) ions at pH 3 whereas that of HPMSP doped mesoporous silica commenced at pH 2.4. Then, the Cd(II) sorption capacity of both silica increased with the increasing of pH and reached its maximum value around pH 4.5. The small amount of extraction at low pH values may be explained by the competition between metal ions and a large amount of protons present in solution for retention on the sorbent. In addition, it is noteworthy that when the extraction experiment was performed at the optimum pH, the Cd(II) sorption capacity of non-doped mesoporous silica and that of HPMSP doped mesoporous silica were similar. These results may be explained by the effect of surface area which played more important role than that of chelating molecules. Indeed, as seen from Table 4.3, the surface area of non-doped mesoporous silica was higher than that of HPMSP doped mesoporous silica.

ii. Extraction of Pb(II)

The effect of pH on the Pb(II) extraction efficiency of both materials was studied in the pH range of 1 to 5. The amounts of Pb(II) extracted by non-doped and HPMSP doped mesoporous silica were illustrated in Figure 4.5.

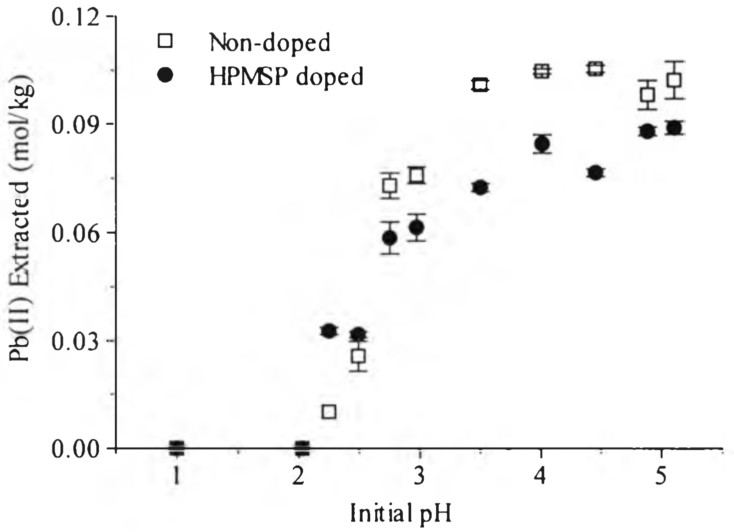

Figure 4.5 Effect of pH on the Pb(II) extraction efficiency of non-doped and HPMSP doped mesoporous silica.

From Figure 4.5, the extraction of Pb(II) by both sorbents was greatly influenced by the pH of metal solution. The Pb(II) extractability was absent at low pH, enhanced with the increasing of pH and attained its maximum value above pH 3.5. These results could be explained by the same reason as described for Cd(II) extraction. However, when the initial pH of solution was higher than 3.5, the Pb(II) extractability of non-doped mesoporous silica was slightly better than that of HPMSP doped mesoporous silica. This meant that the incorporated HPMSP might not play an important role for the extraction of Pb(II). These results were unexpected. In fact, according to literature, several acylpyrazolone ligands can form complex with Pb(II) ions in liquid-liquid extraction [51]. Furthermore, from the research of Tong and co-worker, the HPMSP impregnated microporous silica could extracted the Pb(II) ions. However, the amounts of Pb(II) uptake were only 0.24 mmol/kg [16].

iii. Extraction of Zn(II)

The influence of pH on the Zn(II) extractability of both silica was also investigated by varying the initial pH of metal solution from 1.5 to 5.5. The obtained results were presented in Figure 4.6.

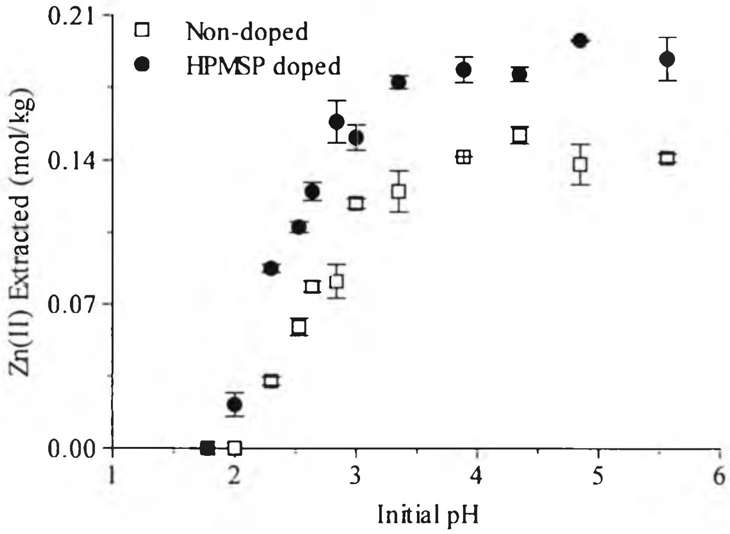

Figure 4.6 Effect of pH on the Zn(II) extractability of non-doped and HPMSP doped mesoporous silica.

According to the results shown in Figure 4.6, the Zn(II) extraction efficiency of both sorbents increased as a function of pH. Interestingly, the HPMSP doped mesoporous silica had higher capacity for the Zn(II) extraction than that of non-doped material through the entire pH range studied. The increase in the Zn(II) sorption capacity of the functionalized silica was probably due to the complex formation between Zn(II) ions and the doping molecules in mesoporous silica.

The comparison of the amounts of each metal extracted by both sorbent at equilibrium had shown the following trend: Zn(II) > Pb(II) > Cd(II). This order was consistent with those reported in the literature [16].

4.3.1.3. Effect of salts present in metal solution

In general, most of the applications of sorbents concern the extraction of metal from various aqueous samples including natural water, seawater and industrial wastewater. These samples contain not only metal ions of interest but also various interferent ions such as Na^+, Mg^{2+}, NO_3^-, OAc^-, Cl^-, etc. which might affect the metal extraction properties of materials. In this study, the influence of diverse ions, in term of different forms of salt, on the metal extraction efficiency of mesoporous silica was thus studied. These salts included $NaNO_3$, NaOAc, NaCl and $Mg(NO_3)_2$ with 0.1 M concentration of each. The extraction experiments were performed on three conditions: in aqueous solution, in metal solution at pH 3 and in metal solution at pH 2. The results of each metal were described below.

i. Extraction of Cd(II)

The amounts of Cd(II) extracted from aqueous solution using non-doped and HPMSP doped mesoporous silica as sorbents were shown in Figure 4.7. The presence of all salts, except NaOAc, in metal solution increased the Cd(II) sorption capacity of both materials. The increasing in Cd(II) sorption capacity was probably due to the replacement of CTA^+ on the surface of silica by Na^+ present in solution. Consequently, the steric hindrance on the surface was decreased and the sorption capacity was promoted. On the contrary, the presence of NaOAc in solution did not significantly affect the Cd(II) extraction capacity of both materials. This result could be explained by the fact that the acetate ions present in metal solution may form complexes with Cd(II) ions to become $Cd(OAc)^+$ [52]. Subsequently, the amounts of free Cd(II) ions in solution was reduced and the Cd(II) extraction was decreased.

For the comparison of Cd(II) extractability of both materials when the metal solution contained different salts, it was found that the sorption capacity of non-doped mesoporous silica and that of HPMSP doped mesoporous silica was not significantly different in all cases except when $Mg(NO_3)_2$ was present in metal solution. For the latter condition, the non-doped mesoporous silica seemed to act as a better sorbent than the HPMSP doped mesoporous silica. These results suggested that the HPMSP molecules in silica might not play any role in the extraction of Cd(II) from aqueous solution.

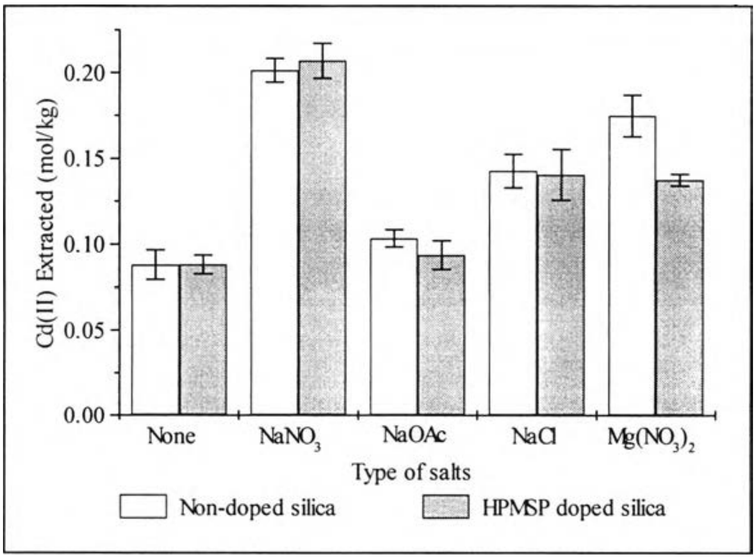

Figure 4.7 Effect of salts present in metal solution on the Cd(II) extraction efficiency of non-doped and HPMSP doped mesoporous silica from aqueous solution.

The influence of salts on the Cd(II) extraction was also studied in acidic medium. The obtained results were depicted in Figure 4.8. The extraction results performed at pH 3 were similar to those carried out earlier in aqueous solution but the Cd(II) extractability of both sorbents were lower. On the contrary, when the experiment was performed at pH 2, the non-doped mesoporous silica completely lost its Cd(II) extraction capacity, whereas the HPMSP doped mesoporous silica could extract some Cd(II) when $NaNO_3$, NaOAc and NaCl were present in solution. The decrease in Cd(II) extractability of both sorbents in this medium was probably due to the competition between a large amounts of proton present in metal solution and metal ions for retention on the sorbent as described previously. Interestingly, among four types of salt used in this study, the highest Cd(II) uptake of modified silica was obtained when NaOAc was present in metal solution. These results could be explained by the fact that when the metal solution contained these ions, the pH was increased due to the hydrolysis of acetate ions and rendered the proper condition for the function of HPMSP molecules incorporated in silica. Consequently, the Cd(II) extractability of HPMSP doped mesoporous silica was promoted.

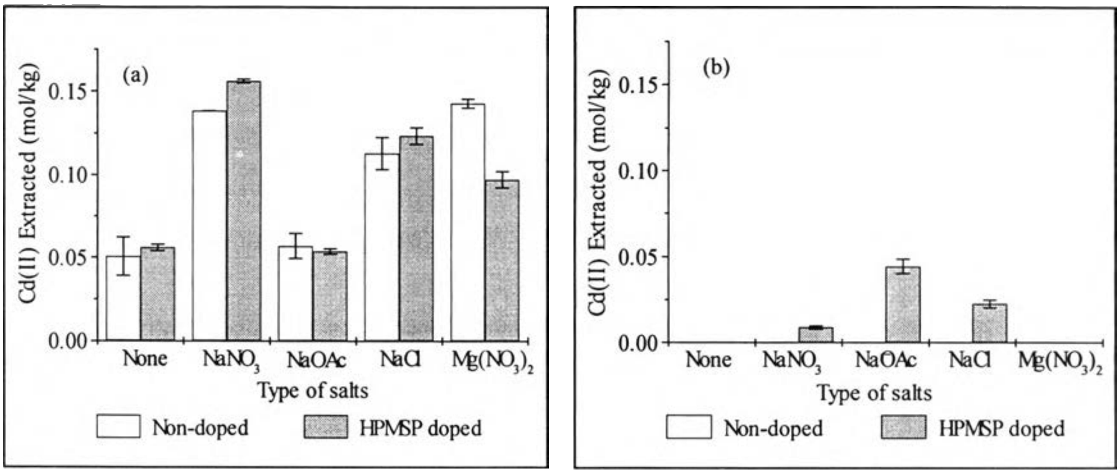

Figure 4.8 Effect of salts present in metal solution on the Cd(II) extraction efficiency of non-doped and HPMSP doped mesoporous silica when the experiments were performed at (a) pH 3 and (b) pH 2.

ii. Extraction of Pb(II)

For the extraction of Pb(II), only $NaNO_3$, NaOAc and $Mg(NO_3)_2$ were the types of salt to study. The NaCl was omitted to avoid the precipitation of $PbCl_2$. The experiments were also performed on three conditions previously stated. The results of Pb(II) uptake when the experiment was performed in aqueous solution were displayed in Figure 4.9.

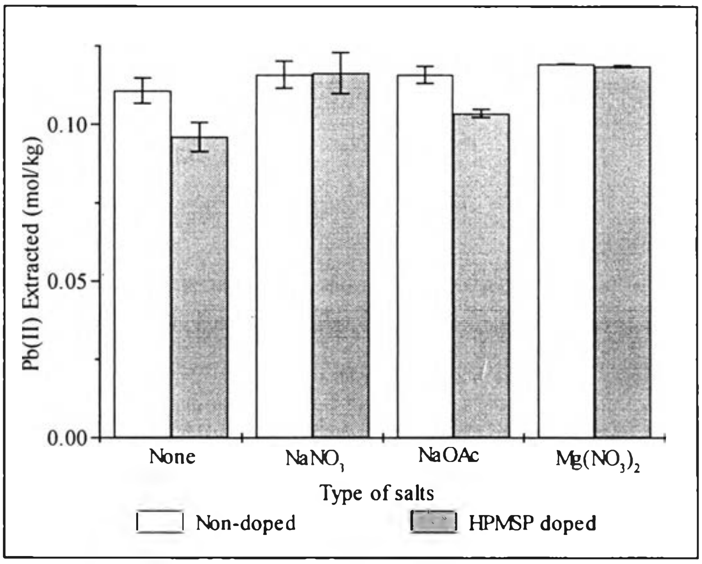

Figure 4.9 Effect of salts present in metal solution on the Pb(II) extraction efficiency of non-doped and HPMSP doped mesoporous silica from aqueous solution.

It is obviously seen from Figure 4.9 that the presence of all salts in metal solution did not significantly affect the Pb(II) extraction capacity of non-doped mesoporous silica, whereas it slightly increased the Pb(II) sorption capacity of HPMSP doped mesoporous silica. However, the amounts of Pb(II) extracted by both materials were not so different, especially when the Pb(II) solution contain $NaNO_3$ or $Mg(NO_3)_2$. This phenomenon indicated that the HPMSP molecules incorporated in silica might not play an important role for the extraction of these ions.

The influence of salts on the Pb(II) extraction were also conducted in acidic medium. The obtained results were shown in Figure 4.10. When the extraction experiments were performed at pH 3, both non-doped and HPMSP doped mesoporous silica had similar manner of Pb(II) extraction. The presence of salts in metal solution enhanced the Pb(II) extraction capacity of both materials. However, the amounts of Pb(II) extracted by both sorbents were not different in most of conditions studied. For the experimental results carried out at pH 2, the presence of all salts in metal solution augmented significantly the Pb(II) extraction, especially for the case of HPMSP doped mesoporous silica. In this condition, the Pb(II) extraction capacity of the modified materials was greater than that of non-doped mesoporous silica. These results might be due to the presence of HPMSP molecules inside the silica as previously stated.

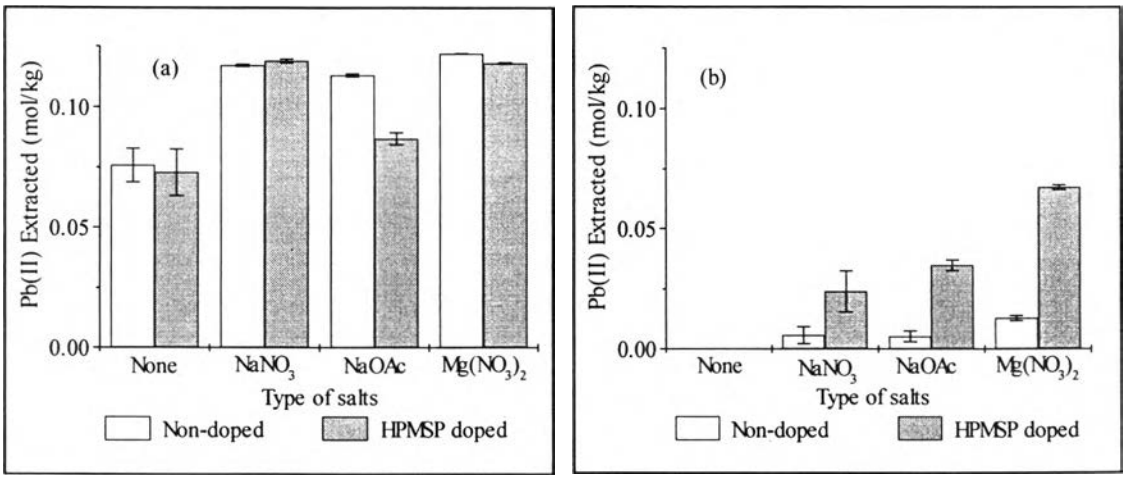

Figure 4.10 Effect of salts present in metal solution on the Pb(II) extraction efficiency of non-doped and HPMSP doped mesoporous silica when the experiments were performed at (a) pH 3 and (b) pH 2.

In addition, apart from using $Pb(NO_3)_2$ as metal source, the influence of salts on the Pb(II) extraction was also performed using $Pb(OAc)_2$ as metal source and the initial pH of lead solution was pH 2. The obtained results were shown in Figure 4.11. The presence of salts, especially $NaNO_3$ and $Mg(NO_3)_2$ in the metal solution augmented significantly the Pb(II) sorption capacity of HPMSP doped mesoporous silica. These results implied that the role of counter ions also had an influence on the Pb(II) extractability of the sorbent. Moreover, it is noteworthy that the modified silica with HPMSP molecules had much more Pb(II) extraction capacity than that of non-doped mesoporous silica. A remarkable difference in these extraction results was mainly caused by the presence of HPMSP molecules in mesoporous silica as stated in section 4.3.1.2.

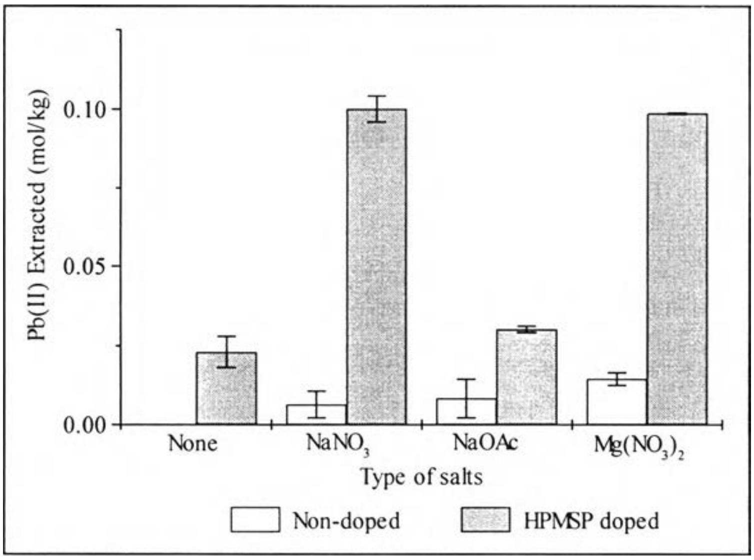

Figure 4.11 Effect of salts present in metal solution on the Pb(II) extractability of non-doped and HPMSP doped mesoporous silica at pH 2 using $Pb(OAc)_2$ as metal source.

iii. Extraction of Zn(II)

Similar experiments was also performed for the study of Zn(II) extraction properties using $Zn(NO_3)_2$ and $Zn(OAc)_2$ as zinc source. The amounts of Zn(II) uptake from aqueous solution were depicted in Figure 4.12. It is obviously seen that the presence of all salts in metal solution increased the Zn(II) extraction capacity of both sorbents using either $Zn(NO_3)_2$ or $Zn(OAc)_2$ as zinc source. These behaviors were the advantage of both materials for the extraction of Zn(II) from samples containing several salts. Furthermore, the HPMSP doped mesoporous silica seemed to had higher Zn(II) sorption capacity than the non-doped mesoporous silica. The advantageous results of the modified silica were probably due to the anchored organic molecules as mentioned previously.

Considering the ability of non-doped and HPMSP doped mesoporous silica to extract Cd(II), Pb(II) and Zn(II), the obtained results showed better extraction properties of both materials towards Zn(II) ions than the other metal ions studied. These results were in accordance with those described in the literature [16].

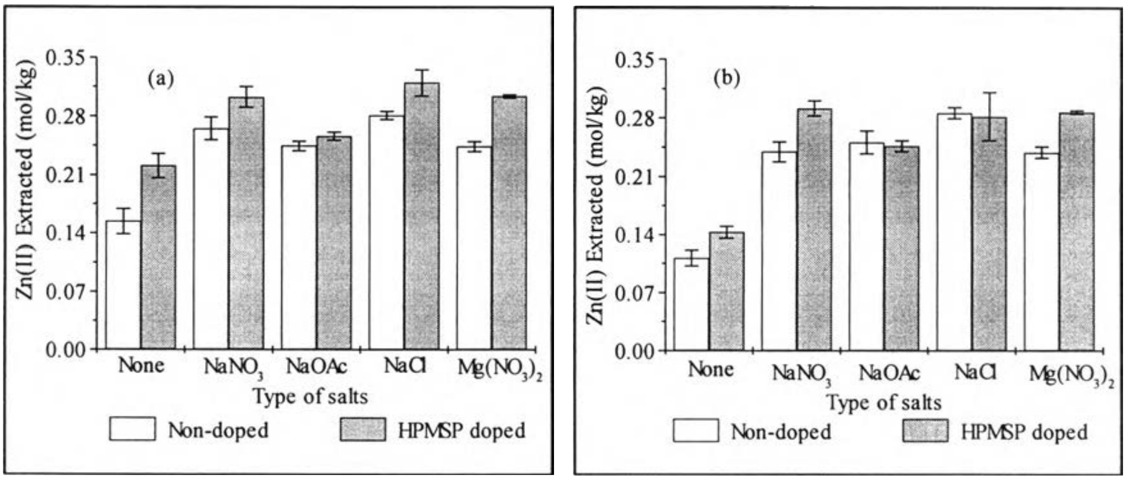

Figure 4.12 Effect of salts present in metal solution on the Zn(II) extraction of non-doped and HPMSP doped mesoporous silica from aqueous solution using (a) $Zn(NO_3)_2$ and (b) $Zn(OAc)_2$ as zinc sources.

The influence of salts on the Zn(II) extraction properties of both materials were also performed at pH 3 and their results were shown in Figure 4.13. By using both $Zn(NO_3)_2$ and $Zn(OAc)_2$ as zinc source, the presence of salts in metal solution enhanced significantly the Zn(II) extraction capacity of both sorbents. Similar results were also observed when the experiments were carried out at pH 2 as illustrated in Figure 4.14. Interestingly, for all conditions studied, the HPMSP doped mesoporous silica had higher Zn(II) extraction capacity than the non-doped mesoporous silica. These results could be explained by the presence of HPMSP molecules in mesoporous silica as previously stated. These extraction behaviors implied that the modified material could be used as a sorbent for the extraction of Zn(II) from solution with low pH values.

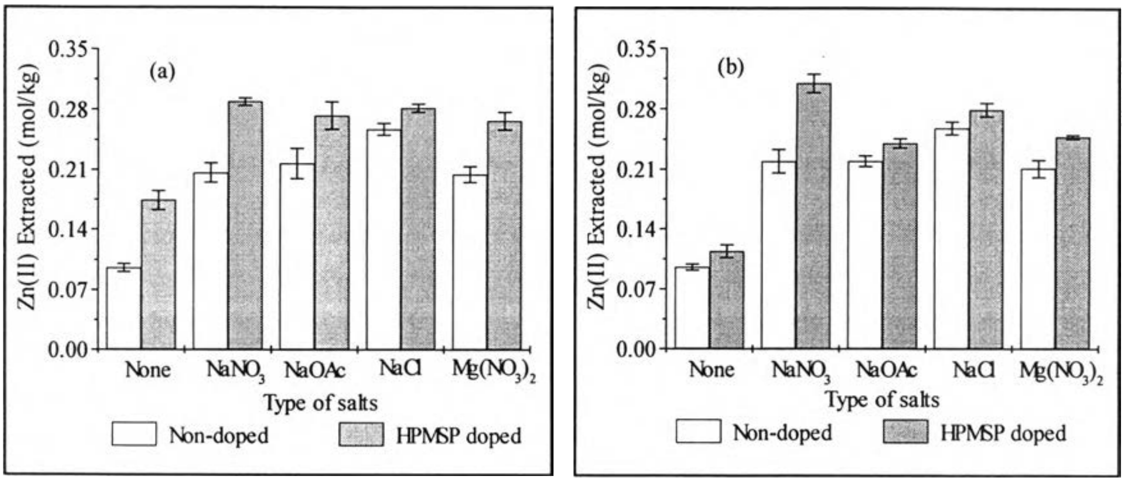

Figure 4.13 Effect of salts present in metal solution on the Zn(II) extraction of non-doped and HPMSP doped mesoporous silica performed at pH 3 and used (a) Zn(NO$_3$)$_2$ and (b) Zn(OAc)$_2$ as zinc sources.

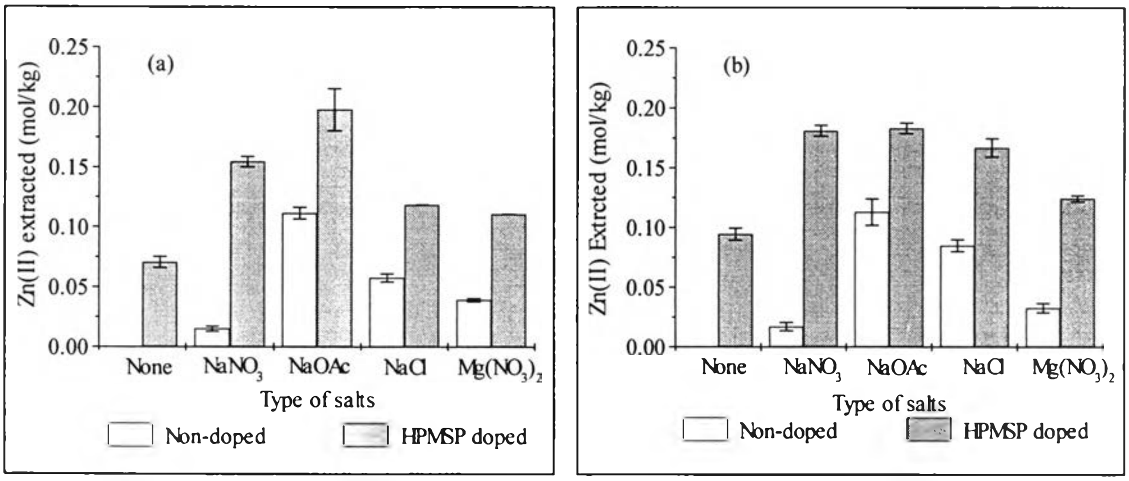

Figure 4.14 Effect of salts present in metal solution on the Zn(II) extraction of non-doped and HPMSP doped mesoporous silica performed at pH 2 and used (a) Zn(NO$_3$)$_2$ and (b) Zn(OAc)$_2$ as zinc sources.

4.3.1.4. Effect of NaNO$_3$ concentration

As shown from the previous section that the presence of NaNO$_3$ in metal solution played an important role in the extraction properties of mesoporous silica. In this section, profound study was thus focused on the influence of NaNO$_3$ concentration on the Zn(II) extractability of both sorbents from aqueous solution using Zn(NO$_3$)$_2$ and Zn(OAc)$_2$ as zinc source. The concentration of NaNO$_3$ was varied from 0 to 0.125 M in case of using Zn(NO$_3$)$_2$ as zinc source and it was varied from 0 to 0.1 M when Zn(OAc)$_2$ was used. The obtained results were displayed in Figure 4.15.

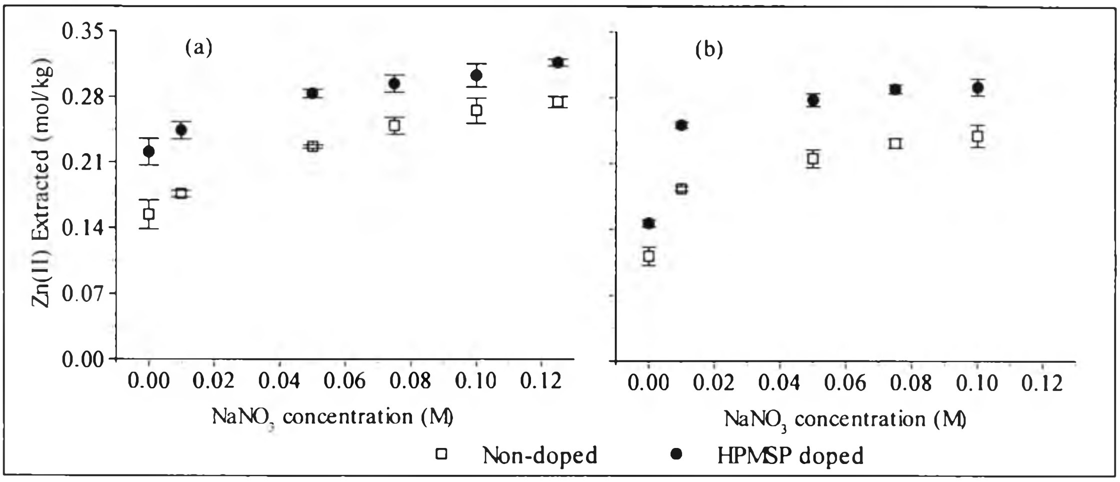

Figure 4.15 Effect of $NaNO_3$ concentration on the extraction of Zn(II) by non-doped and HPMSP doped mesoporous silica using (a) $Zn(NO_3)_2$ and (b) $Zn(OAc)_2$ as zinc sources.

From Figure 4.15, the Zn(II) extractability of both materials seemed to be increased with the increasing of $NaNO_3$ concentration when either $Zn(NO_3)_2$ or $Zn(OAc)_2$ was used as zinc sources. In both cases and for both sorbents, the maximum uptake of Zn(II) was obtained when the concentration of $NaNO_3$ was equal or more than 0.075 M. The increase in Zn(II) extractability when the metal solution contained the sodium salt may be explained by the replacement of CTA^+ on the surface of silica by Na^+ as described previously. However, when the concentration of $NaNO_3$ was equal or more than 0.075 M, the Zn(II) extractability of both sorbents remained constant. These phenomena were probably due to the complete replacement of all CTA^+. Consequently, the much more increasing of $NaNO_3$ concentration was in vain. Considering the Zn(II) extraction ability of both sorbents, the HPMSP doped mesoporous silica seemed to had better extractability than the non-doped mesoporous silica through the entire concentrations studied. These higher extracted values of the modified silica might be attributed to the existence of HPMSP molecules in mesoporous silica.

4.3.1.5. Comparison with other sorbents

The maximum sorption capacity of HPMSP doped mesoporous silica in this work was listed and compared with those of other functionalized silica as summarized in Table 4.6. From this table, the modified sorbent had shown better or comparable capacity values to most of other sorbents used for the enrichment of Cd(II), Pb(II) and Zn(II).

Table 4.6 Comparison of sorption capacities (mol/kg).

Ligand	Functionalization method	Metal ions		
		Cd(II)	Pb(II)	Zn(II)
1-Phenyl-3-methyl-4-stearoyl-5-pyrazolone (this work)	Doping	0.2070	0.1186	0.3199
Dithizone [53]	Grafting	0.0350	0.0400	0.0200
Purpurogallin [54]	Grafting	0.0400	0.0300	0.1199
1-(2-Pyridylazo)-2-naphthol [11]	Doping	0.0440		
Resacetophenone [55]	Grafting	0.0528	0.0620	0.1881
1,8-Dihydroxyantraquinone [56]	Grafting	0.0702	0.0764	0.1804
1-(2-Thiazolylazo)-2-naphthol [46]	Doping			0.0350
Salicylaldoxime [57]	Grafting			0.0400
2,5-Dimercapto-1,3,4-thiadiazole [58]	Grafting	0.1480	0.1530	0.1440

4.3.2. Column method

4.3.2.1. Reproducibility of material

One of the crucial factors to indicate how good of material for acting as a sorbent is the reproducibility. In this work, the reproducibility of HPMSP doped mesoporous silica towards the metal extraction was investigated using Cu(II) as a metal ion. This element was chosen due to the strong selectivity between it and the doping molecules. The experiments were performed by loading 300 mL of 20 ppm or 40 ppm of Cu(II) solution into the column. The medium of the metal solution was $5x10^{-3}$ M $NaNO_3$ at pH 2. For each concentration, the experiments were repeated sixteen times. The average amounts of Cu(II) extracted were found to be 0.0356 ± 0.0025 mol/kg and 0.0484 ± 0.0032 mol/kg when the initial concentration of Cu(II) was 20 ppm and 40 ppm, respectively. The results obtained on 16 replicate measurements revealed a RSD of 7% for both initial concentrations (see Appendix E for further details). The prepared sorbent was subjected to further column capacity determination.

4.3.2.2. Optimization of metal desorption

The reusability of materials is one of the major aims for most of researches concerning the development of sorbent because it can reduce potentially the high cost of separation, sample preparation and the problem of waste disposal. However, prior to reuse the sorbents, all the adsorbed substances have to be withdrawn from such materials. In this work, the desorption properties of HPMSP doped mesoporous silica was then optimized using an experimental design namely 2^3 factorial design. The amounts of Cu(II) loaded into the column, the concentration of HNO_3 as an eluent and the volume of this acid are three experimental parameters to be optimized. Each variable contained two levels and each experimental run was performed in triplicate. The experimental variables, codes, levels, design matrix and the results were summarized in Table 4.7.

Table 4.7 Experimental variables, levels, design matrix and results (amounts of Cu(II) desorped) in the 2^3 factorial design for Cu(II) desorption.

Variables	Codes	Level	
		Low (-)	High (+)
Amount of Cu(II) loaded (μmol)	A	94.4	188.8
Concentration of HNO_3 (M)	E	1	3
Volume of HNO_3 (mL)	V	20	40

Run	A	E	V	Amounts of Cu(II) desorped (mol/kg)
1	-	-	-	0.0309 ± 0.0012
2	+	-	-	0.0327 ± 0.0009
3	-	+	-	0.0343 ± 0.0039
4	+	+	-	0.0402 ± 0.0003
5	-	-	+	0.0255 ± 0.0030
6	+	-	+	0.0346 ± 0.0016
7	-	+	+	0.0335 ± 0.0017
8	+	+	+	0.0382 ± 0.0021

From the amounts of Cu(II) desorbed, the influence of each variable and their interaction could be determined by the calculation of estimated effects (see Appendix D). The results and their standard errors (SE) were presented in Table 4.8.

Table 4.8 The estimated effects of the variables and their interaction.

Variables/Interactions	Estimated effects (x 10^{-3}) ± SE (x 10^{-3})
Main	
Amounts of Cu(II) loaded, A	5.375 ± 0.874
Concentration of HNO_3, E	5.625 ± 0.874
Volume of HNO_3, V	-1.575 ± 0.874
Two-factor interaction	
AE	-0.075 ± 0.874
AV	1.525 ± 0.874
EV	0.175 ± 0.874
Three-factor interaction	
AEV	-2.125 ± 0.874

In general, the influence of factors or interactions is considered as significance if their estimated effect is higher than the standard error. Thus, from Table 4.8, the significant factors were the amounts of Cu(II) loaded (A), the concentration of HNO_3 (E), the volume of HNO_3 (V), the AV interaction and the three factor interaction (AEV). However, among these significant factors, the concentration of nitric acid and the amounts of Cu(II) loaded seemed to be much more important than other variables/interactions due to their higher estimated effects. In addition, the positive sign of these estimated effects indicated the increasing of Cu(II) desorption efficiency when high concentration of HNO_3 and large amounts of Cu(II) loaded were used.

The analysis of variance (ANOVA) was also performed to identify the significant factor affecting the Cu(II) desorption of the sorbent (Table 4.9). The significant factor was verified by considering the F-ratio and the P-value. The influence of factor or interaction is considered as significant if the F-ratio is higher than F-critical value. Moreover, when the P-value is less than 0.05, the factor has a statistically significant effect at the 95% confidence level. As could be seen from Table 4.9, two main factors including the concentration of HNO_3 and the amounts of

Cu(II) loaded were found to be significant. Also, the three-factor interaction was significant.

Table 4.9 Analysis of variance for the Cu(II) desorption efficiency of HPMSP doped mesoporous silica.

Source of variance	Sum of squares (x 10^{-6})	Degree of freedom	Mean squares (x 10^{-6})	F-ratios	P-values
A	173.34	1	173.34	48.89	<0.0001
E	189.84	1	189.84	53.54	<0.0001
V	14.88	1	14.88	4.20	0.0573
AE	0.03	1	0.03	0.01	0.9279
AV	13.9	1	13.9	3.93	0.0647
EV	0.18	1	0.18	0.05	0.8246
AEV	27.09	1	27.09	7.64	0.0138
Error	56.73	16	3.55		
Total	494.25	23			

Sum of square = N x (estimated effect)^2/4, Mean square = sum of square/degree of freedom, F-ratio = mean square of factor/mean square of error, $F_{1,16} = 6.115$

In addition, the significant factor could also be confirmed from the normal probability plot of estimated effect as displayed in Figure 4.16. When the factors are not laid along the straight line, they are significant. As seen from Figure 4.16, three points including the amounts of Cu(II) loaded, the concentration of HNO_3 and the three-factor interaction were thus significant. These results were consistent with the results obtained from the analysis of variance (ANOVA).

In conclusion, the significant factor affecting the Cu(II) desorption efficiency of HPMSP doped mesoporous silica was the concentration of HNO_3, the amounts of Cu(II) loaded and the three-factor interaction. Thus, to obtained the high Cu(II) desorption efficiency of the sorbent, the optimum condition used for the desorption should be the high concentration of nitric acid and the large amounts of Cu(II) loaded.

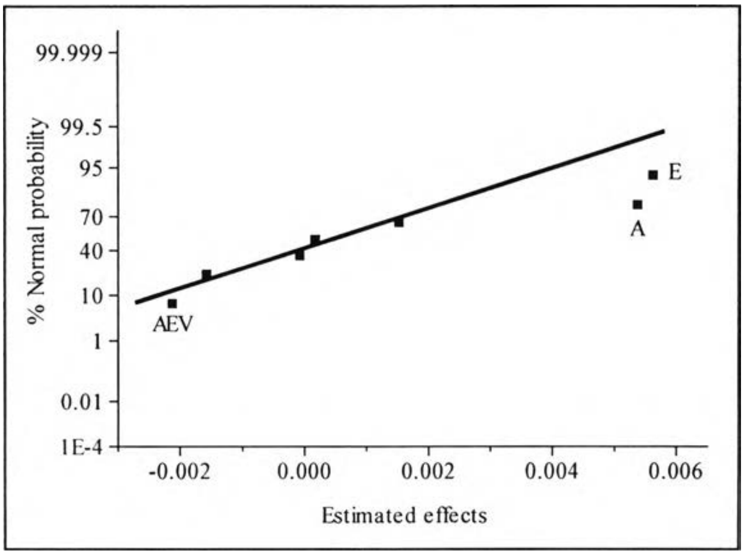

Figure 4.16 The normal probability plot of estimated effect.

4.3.2.3. Column capacity

The capacity of materials is an important factor that determines how many of metal ions could be extracted by sorbent. In this work, the column capacity of HPMSP doped mesoporous silica was evaluated using Cu(II) solution in $5x10^{-3}$ M $NaNO_3$ medium at pH 2 and the range of Cu(II) concentration was from 50 ppm to 300 ppm. The results plotted between the initial Cu(II) concentration and the amounts of Cu(II) extracted were displayed in Figure 4.17.

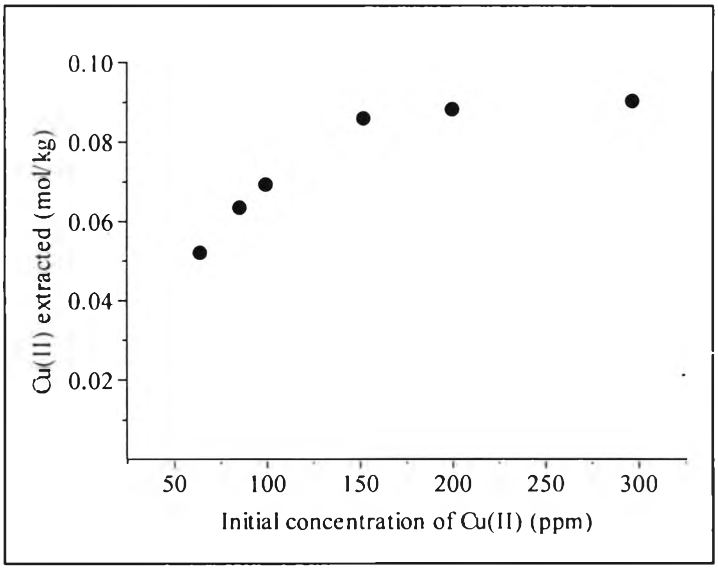

Figure 4.17 The amounts of Cu(II) extracted by HPMSP doped mesoporous silica using column method as a function of initial concentration of Cu(II).

From Figure 4.17, the Cu(II) extractability of HPMSP doped mesoporous silica seemed to be increased with the increasing of initial concentration of these metal ions. To investigate the adsorption isotherm type, the amounts of Cu(II) sorption (N_f) were plotted against the Cu(II) concentration at equilibrium (C_e) as shown in Figure 4.18 (a). From this figure, the obtained profile followed the Langmuir model. Considering the plot of C_e/N_f versus C_e as shown in Figure 4.18 (b), the straight line was obtained with a slope of $1/N_S$. The equilibrium constant (K_L) calculated from the intercept of the langmuir plot was found to be 0.0802 mL/mg. Also, the maximum extraction capacity (N_s) determined from the slope was found to be 6.34 mg/g (0.0998 mol/kg). This value was superior to Cu(II) extraction capacity of other functionalized sorbents reported elsewhere (i.e. 0.0780 mol/kg for the Cu(II) sorption capacity of zirconium phosphate functionalized silica and 0.0063 mol/kg for Schiff's base modified octadecyl silica membrane disks) [59,60].

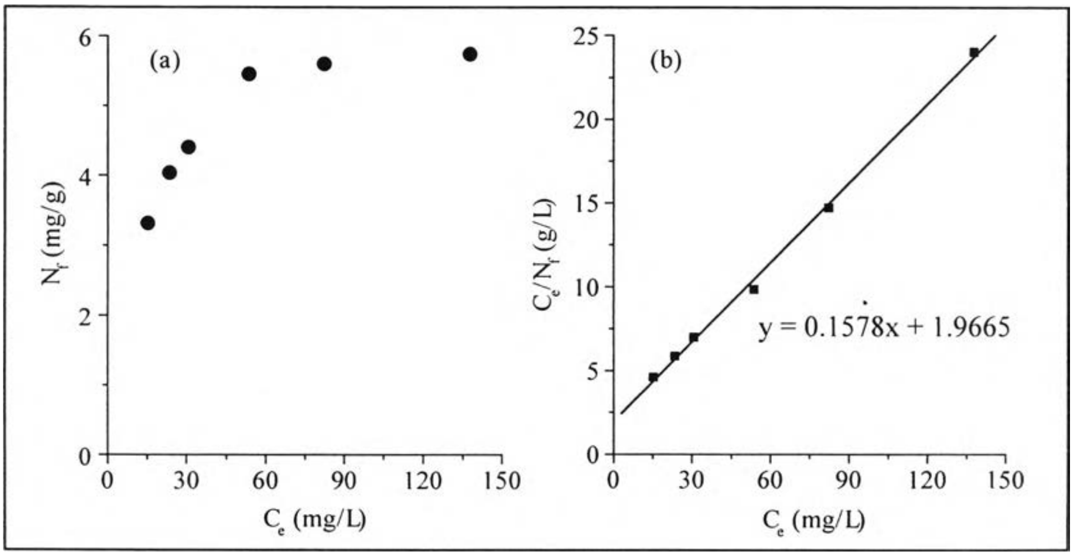

Figure 4.18 (a) Adsorption isotherms of Cu(II) on HPMSP doped mesoporous silica and (b) linearization of the adsorption isotherm.

4.3.2.4. The effect of sample volume

For the preconcentration of trace elements in natural water samples, the method used should have high preconcentration factor. Sample volume is one of the crucial parameters which has an effect on the preconcentration efficiency. In this work, the effect of sample volume on the Cu(II) desorption efficiency of HPMSP doped mesoporous silica was investigated by loading different volumes of Cu(II) solution (i.e. 100, 200, 300 and 800 mL) to the column. The extraction medium used was 5×10^{-3} M $NaNO_3$ at pH 2 and 20 mL of 3 M HNO_3 was used as eluent through the desorption process. The obtained results were reported in Table 4.10.

Table 4.10 Effect of sample volume on the Cu(II) desorption efficiency of HPMSP doped mesoporous silica.

Volume of Cu(II) solution (mL)	Volume of eluent (mL)	Preconcentration factor	Recovery (%)
100	20	5	96
200	20	10	95
300	20	15	92
800	20	40	94

All results displayed in Table 4.10 showed relatively high recovery of Cu(II) which suggested that the metal sample volume did not have much effect on the Cu(II) desorption efficiency of this sorbent. The highest preconcentration factor in this study was found to be 40. Though this value could be augmented by loading much more metal solution volume, this increase in volume experiment was not carried out due to an inappropriate time of analysis.

4.3.2.5. Reusability of material

The reusability of materials is one of the major aims for most researches concerning the development of sorbent because it can reduce the problem of waste disposal and the cost of sample separation and preparation. To demonstrate the potential reusability of HPMSP doped mesoporous silica, this sorbent was subjected to several loading with 60 mL of 150 ppm of Cu(II) solution in 5×10^{-3} M $NaNO_3$ medium at pH 2 and eluting with 20 mL of 3 M HNO_3 as desorption agent. The Cu(II) adsorption-desorption cycle was repeated three times. The obtained results were displayed in Table 4.11. As seen, the adsorption capacity of the sorbent did not change after three cycles of adsorption and desorption. These results showed that these modified materials were suitable for repeated use.

Table 4.11 The Cu(II) adsorbed and desorbed on HPMSP doped mesoporous silica, using 20 mL of 3 M HNO_3 as eluent in the column method.

Cycle	Amounts of Cu(II)	
	Adsorption (mol/kg)	Desorption (mol/kg)
1	0.0789	0.0780
2	0.0782	0.0747
3	0.0685	0.0639

4.4. Application of HPMSP doped mesoporous silica to metal extraction from food sample

4.4.1. The determination of metal ions contaminated in seafood sample

The determination of metal ions at trace level is of prime importance to the context of environmental protection, food safety and agricultural production. However, the direct determination of trace metals is limited due to their low concentration and matrix interferences. Therefore, a preconcentration and/or separation of trace elements from matrix is frequently required in order to achieve an appropriate level of sensitivity. One of the most convenient approach to meeting this need is solid-phase extraction. Thus, one of the purpose of this work is to demonstrate the feasibility of using HPMSP doped mesoporous silica as a sorbent for the extraction of metal contaminated in food. Two varieties of food such as mussel and fish were used. The preliminary experiments were performed to determine the types and the amounts of metal ions existing in both samples. Prior to analysis, the samples need to be digested to destroy the existing matrix.

The first microwave digestion experiment was carried out on mussel sample. This digestion method was chosen due to its low consumption of reagent and rapidity of digestion time. When the digestion process was completed, the digested sample was subjected to metal analysis. Due to their association with pollution and toxicity problems, six metal ions including Cd(II), Co(II), Cu(II), Ni(II), Pb(II) and Zn(II) became the metal of interest in this work. The metal determination results had shown that the digested mussel sample contained only Ni(II) and Zn(II). On the contrary, the extents of other metal ions could not be determined since their quantities were below the detection limit of AAS. The considerably low signal may be explained by the fact that the Cd(II), Co(II), Cu(II) and Pb(II) ions were totally absent from the mussel sample or the initial quantity of mussel sample used for the digestion was relatively too small. To know the certain cause, the old microwave digestion was thus replaced by the wet digestion method in order to work with much more amounts of sample. For the latter method of sample preparation, both mussel and fish were served as sample matrix. The experiment was repeated four times and four more metal ions such as Ca(II), Fe(III), Mg(II) and Mn(II) were also determined. The obtained results were summarized in Table 4.12. As seen, both mussel and fish samples contained Ca(II) and Mg(II) in a relatively high quantity comparing to other metal ions. The high amount of such ions was expected since these ions are among the most abundant ions in the sea [61]. For the metals whose amounts were below the detection limit of AAS, it may be explained by the incomplete destruction of matrix. Consequently, the release of metal ions to the solution was limited.

In addition, the determination of metal ions in the blank sample had shown the absence of all metals of interest. This result indicated that the contamination of metal ions during the sample preparation did not occur.

Table 4.12 The amounts of metals contained in mussel and fish samples determined by AAS after wet digestion process.

Types of metal	Amounts of metal (µg/g)		Types of metal	Amounts of metal (µg/g)	
	Mussel sample	Fish sample		Mussel sample	Fish sample
Ca(II)	393.07 ± 43.93	158.28 ± 54.32	Mg(II)	575.23 ± 6.12	237.30 ± 35.90
Cd(II)	BDL	BDL	Mn(II)	9.59 ± 0.48	BDL
Co(II)	BDL	BDL	Ni(II)	11.44 ± 4.44	12.19 ± 5.42
Cu(II)	0.96 ± 0.16	BDL	Pb(II)	1.08 ± 0.80	0.96 ± 0.00
Fe(III)	37.90 ± 5.10	12.21 ± 0.14	Zn(II)	10.99 ± 2.69	6.79 ± 1.38

BDL Below the detection limit of AAS

4.4.2. The influence of acetate buffer on the metal extraction properties of HPMSP doped mesoporous silica

As remarked from the digestion process that the final pH of the digested solution after dilution with deionized water was less than 0. Consequently, at this pH, the capacity of all materials is absent. To ameliorate the extraction efficiency of the sorbents, the dilution solvent is thus replaced by the acetate buffer pH 4.5. Therefore, prior to use the HPMSP doped mesoporous silica for the extraction of metal in seafood sample, the sorption behaviors of this sorbent in acetate medium had to be clarified. In this study, the extraction of each metal by the HPMSP modified material was performed in acetate buffer medium at pH 4.5. Six target metals including Cd(II), Co(II), Cu(II), Ni(II), Pb(II) and Zn(II) were determined separately. The concentration of each metal was 200 ppm. The experiments were performed in triplicate and the obtained results were displayed in Table 4.13.

Table 4.13 The amounts of metal ions extracted by HPMSP doped mesoporous silica from acetate buffer medium at pH 4.5.

Types of metal ions	Amount of metal extracted (mol/kg)
Cd(II)	0.0000 ± 0.0000
Co(II)	0.1659 ± 0.0025
Cu(II)	0.1806 ± 0.0049
Ni(II)	0.1528 ± 0.0109
Pb(II)	0.0000 ± 0.0000
Zn(II)	0.1308 ± 0.0147

From that table, among six metal ions studied, the Cd(II) and Pb(II) could not be extracted by this sorbent from the condition studied. These results conflicted with the aforementioned extraction studies (section 4.3.1) and those reported in the literature review [14, 16, 51]. Therefore, the acetate buffer pH 4.5 used in this work might not be an inappropriate medium for the extraction of Cd(II) and Pb(II). For other metal ions, the highest metal extractability of the sorbent was observed for Cu(II), followed by Co(II), Ni(II) and Zn(II). These results were not strange to us since the HPMSP molecules were favorable for the extraction of Cu(II) [16, 62].

4.4.3. The metal extraction of HPMSP doped mesoporous silica from mixture solution in acetate buffer medium

In general, environmental samples including natural water, seawater and wastewater contain various metal ions such as Cd(II), Co(II), Cu(II), Mn(II), Ni(II), Zn(II), etc. [63-64]. Thus, the comprehension of metal extraction properties of sorbent from mixture solution of several metal ions is often necessary. In this work, the metal extraction properties of HPMSP doped mesoporous silica from mixture solution were investigated using acetate buffer pH 4.5 as a medium. Two mixtures of metal ion solution including Cd(II)/Cu(II)/Pb(II)/Zn(II) and Co(II)/Cu(II)/Ni(II)/Zn(II) were used. The mixtures contain 50 ppm concentration of each metal. The results of each extracted ion expressed in mol/kg were compiled in Table 4.14.

Table 4.14 The amounts of each extracted metal ion from mixture solution in acetate buffer medium at pH 4.5.

Mixture solution	Type of metal ions	Amounts of metal extracted (mol/kg)
Mixture I	Cd(II)	0.0000 ± 0.0000
	Cu(II)	0.0916 ± 0.0016
	Pb(II)	0.0000 ± 0.0000
	Zn(II)	0.0223 ± 0.0017
Mixture II	Co(II)	0.0315 ± 0.0008
	Cu(II)	0.0847 ± 0.0023
	Ni(II)	0.0367 ± 0.0008
	Zn(II)	0.0235 ± 0.0010

From Table 4.14, the lack of Cd(II) and Pb(II) extraction ability of the HPMSP doped mesoporous silica form mixture solution I was still observed. These results confirmed the inappropriate extraction condition of the acetate buffer medium for both metal ions. For other metal ions, the remarkable selectivity of the modified silica was observed for the extraction of Cu(II). Considering the metal extraction from mixture solution II, the metal extractability of this sorbent was found to be in this sequence: Cu(II) >> Ni(II) ≅ Co(II) > Zn(II). These results clearly indicated the strong selectivity of HPMSP doped mesoporous silica to the Cu(II) extraction. This great selectivity of the modified sorbent is of benefit to the extraction of this ion from samples containing other metal ions.

4.4.4. The extraction of metal in seafood matrix

From section 4.4.1, the amounts of metal ions contained in mussel and fish samples were relatively low. To demonstrate the potential application of HPMSP doped mesoporous silica for the extraction of metal from the seafood samples, the known amount of metal ions was thus spiked into the sample prior to the digestion experiment. Two series of metal ion mixture including Cd(II)/Cu(II)/Pb(II)/Zn(II) and Co(II)/Cu(II)/Ni(II)/Zn(II) were used as spiking ions. The mixtures contained 500 ppm concentration of each metal. In each digestion batch, 10 mL of the metal mixture were spiked into the samples. Then the spiked samples were brought to the wet digestion (see section 3.3.4). To complete the digestion process, the acetate buffer pH 4.5 was used as a dilution solvent to make a final concentration of 50 ppm of each metal in both digested samples. The blank which contained neither mussel nor fish was also digested in a similar manner to use for comparison. The recovery of metal ions in the blank and both samples was reported in Table 4.15.

Table 4.15 The percent recovery results for metal ions added to the blank, mussel and fish samples.

Sample matrix	Mixture I				Mixture II			
	Cd(II)	Cu(II)	Pb(II)	Zn(II)	Co(II)	Cu(II)	Ni(II)	Zn(II)
Blank	98.6	95.4	101.6	98.6	99.2	98.2	99.6	95.2
Mussel	99.0	98.8	92.0	99.6	102.2	101.6	99.0	97.6
Fish	96.0	98.0	105.4	99.2	101.0	100.2	99.0	97.6

As seen from Table 4.15, the recovery of all metal ions in every digested sample was close to 100%. It could thus be concluded that there was no loss of metals of interest during the sample preparation. Furthermore, the sample matrices did not present any effect on the recovery results.

All digested samples were then applied to the determination of the sample matrix on the metal extraction properties of HPMSP doped mesoporous silica. The obtained results based on the average of three replicates were tabulated in Table 4.16.

Table 4.16 Effect of sample matrix on the amounts of metal extracted by HPMSP doped mesoporous silica in each mixture.

Mixture I:

Sample matrix	Amounts of metal extracted (mol/kg)			
	Cd(II)	Cu(II)	Pb(II)	Zn(II)
Blank	0.0000 ± 0.0000	0.0936 ± 0.0010	0.0000 ± 0.0000	0.0353 ± 0.0007
Mussel	0.0000 ± 0.0000	0.0943 ± 0.0005	0.0000 ± 0.0000	0.0087 ± 0.0016
Fish	0.0000 ± 0.0000	0.0958 ± 0.0002	0.0000 ± 0.0000	0.0411 ± 0.0062

Mixture II:

Sample matrix	Amounts of metal extracted (mol/kg)			
	Co(II)	Cu(II)	Ni(II)	Zn(II)
Blank	0.0266 ± 0.0015	0.0962 ± 0.0001	0.0441 ± 0.0017	0.0133 ± 0.0012
Mussel	0.0105 ± 0.0014	0.0992 ± 0.0001	0.0215 ± 0.0022	0.0068 ± 0.0006
Fish	0.0280 ± 0.0007	0.0977 ± 0.0001	0.0430 ± 0.0001	0.0143 ± 0.0003

From Table 4.16, the results obtained from the blank were similar to those observed from section 4.4.3. The Cd(II) and Pb(II) ions were not still extracted by the modified silica. On the contrary, this sorbent could extract other metal ions and the highest uptake was observed for the Cu(II). Considering the effect of sample matrix, only the mussel matrix had found to influence the extraction properties of the modified material, especially for the extraction of Co(II), Ni(II) and Zn(II). However, this matrix did not have any effect on the extraction of Cu(II). On the contrary, the impact of fish matrix on the metal extraction properties of the sorbent was absent since the amounts of metal extracted from this sample matrix were close to those observed from the blank. In conclusion, this HPMSP modified silica seemed to be a suitable candidate for metal extraction from fish sample.