



ISSN: 0067-2904

## Synthesis, Characterization and Application of Calcium Ion-Imprinted Polymeric Solid-Phase Extraction and Pre-Concentration in Aqueous Solutions by Packed-Bed Columns

Ghaidaa sabry hassoon\*, Yehya Kamal Al-Bayati

Department of Chemistry, College of Science, University of Baghdad, Baghdad, Iraq

Received: 28/6/2022 Accepted: 25/12/2022 Published: 30/11/2023

### Abstract

In this work, calcium ions were determined by the addition to the allyl chloride monomer resulting from bulk polymerization formation. To acquire the highest adsorption capacity, molar ratios of the template, monomer, and cross-linking agent, as well as solvents and multiple monomers were investigated. Scanning Electron Microscopy (SEM) and Fourier Transform Infrared Spectroscopy (FTIR) were used to analyze the calcium ion polymer. The elution of calcium had a small effect on the surfaces of the three-dimensional network structure. Calcium (II) ions were successfully eluted using a mixture of methanol and acetic acid. The calcium adsorption capacities were 3.8145 and 9.01773 mol/g ( $Q_{max}$ ), respectively. A Langmuir isotherm model follows calcium adsorption. Solid-phase extraction (SPE) syringes packed with ionic imprinted polymers (IIPs) were used to selectively separate and preconcentration the calcium (II) ion from aqueous solutions to determine the calcium ion by flame atomic absorption spectroscopy (FAAS).

**Keywords:** Molecularly imprinted polymer; calcium ion; allyl chloride, monomers.

تخليق وتشخيص وتطبيق استخلاص الطور الصلب البوليمري المطبوع بأيون الكالسيوم والتركيز المسبق في المحاليل المائية بواسطة اعمدة طبقة معبأة

غيداء صبري حسون\* ، يحيى كمال البياتي

قسم الكيمياء كلية العلوم، جامعة بغداد، بغداد ، العراق

### الخلاصة

في هذا العمل، تم تحديد أيونات الكالسيوم من خلال إضافة مونومر كلوريد الأليل الناتج عن تكوين البلمرة الكتلية. للحصول على أعلى سعة امتصاص، تم فحص النسب المولية لل قالب، والمونومر، وعامل الربط المتبادل، وكذلك المذيبات والمونومرات المتعددة. تم استخدام المسح المجهر الإلكتروني (SEM) و Fourier Transform Infrared Spectroscopy (FTIR) لتحليل بوليمر أيون الكالسيوم. كان لشطف الكالسيوم تأثير ضئيل على أسطح بنية الشبكة ثلاثية الأبعاد. تمت تصفية أيونات الكالسيوم (II) بنجاح باستخدام خليط من الميثانول وحمض الخليك. كانت سعات امتصاص الكالسيوم 3.8145 و 9.01773 على التوالي. يتبع امتزاز الكالسيوم من نوع لانكماير تم استخدام حقنة للاستخلاص بالطور الصلب (SPE) المعبأة بالبوليمرات

\* Email: [gadaasabree@gmail.com](mailto:gadaasabree@gmail.com)

الأيونية المطبوعة (IIPs) للفصل الانتقائي والتركيز والتقدير للأيون الكالسيوم (II) من المحاليل المائية بطريقة مطيافية الامتصاص الذري باللهب (FAAS).

## 1. Introduction

Calcium is a metallic element that comprises more than 3% of the earth's crust, ranking fifth in abundance. It is found in leaves, bones, teeth, shells, etc. It has never been found in nature alone, and uncombined calcium seems to be silvery [1]. Calcium is an essential component of human and animal bodies because it protects the bone system and serves as a regulatory ion both within and outside the cell. The calcium ion concentration in aqueous media can be measured for a range of applications, including industrial and domestic water hardness management, food quality assessment, and medical diagnosis [2]. When the calcium level of water exceeds  $4 \times 10^{-3} \text{ mol L}^{-1}$ , a plethora of industrial difficulties emerge, including scale development [3]. The activity of calcium ions is widely monitored in medical diagnosis and treatment. Calcium ion levels that rise or fall outside the normal range can have serious consequences for your health (the normal range:  $2.2\text{-}2.5 \times 10^{-3} \text{ mol.L}^{-1}$ ) [4]. Bulk polymerization is the most straightforward way to create pure polymer forms [4]. The ion-imprinting process consists of three steps: (i) template (metal ions) complexation with a polymerizable ligand; (ii) polymerization of this complex; and (iii) template removal after polymerization [5]. The specificity of the ligand, the coordination geometry, and the coordination number of the ions, as well as their charges and sizes, all have an influence on the selectivity of a polymeric adsorbent in the ion imprinting process [6]. The template can be delivered into the system in a variety of ways, including standing alone or being bonded to a surface, resulting in 3D or 2D imprinting environments that respect polymerization. This could be covalent, non-covalent, or semi-covalent in nature [7]. Non-covalent imprinting is by far the most prevalent approach due to its ease of production and the vast range of monomers available [8]. By virtue of their existence, they are required for molecular interactions. Acceptance procedures are widely employed since they are considered the most efficient and successful method for molecularly imprinted polymer (MIP) synthesis. The researchers are currently working on creating a method for selective preconcentration of sorbents utilized in solid-phase extraction (SPE) [9]. Wastewater or river water is examples of complicated matrices. SPE is a more straightforward, quick, and cost-effective method of extraction that is also environmentally friendly [5, 10]. The most significant issue is the use of standard stationery stacked in SPE columns. With the retention phase's low selectivity mechanism, it's possible to achieve a desired level of selectivity [10, 11]. Here, we investigate the selective separation and preconcentration of the calcium (II) ions from aqueous solutions by the addition of an allyl chloride monomer, resulting in bulk polymerization formation, to determine the calcium ion by flame atomic absorption spectroscopy (FAAS).

## 2. Experimental part

### 2.1. Materials and method

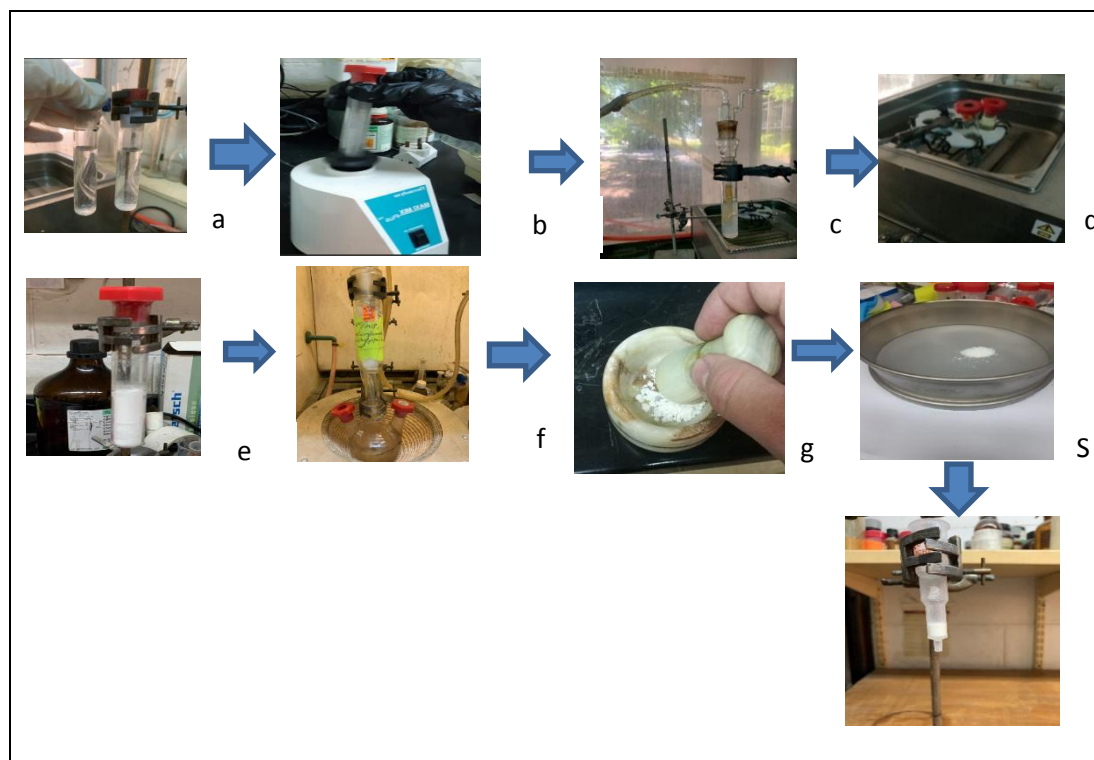
Calcium chloride dihydrate (99.9%), allyl chloride (99.9%), ethylene glycol methacrylate (EGDMA) (99.9%), benzoyl peroxide were purchased from Sigma Aldrich (St. Louis, MO, USA, [www.sigma-aldrich.com](http://www.sigma-aldrich.com)), methanol, nitrogen gas and acetic acid were purchased from Merck (Darmstadt, Germany).

### 2.2. MIP procedure

Preparation of ionic imprinted polymer: For the preparation of the number one calcium ionic imprinted polymer (calcium-IIP), calcium chloride dihydrate (147 mg, 1 mmol) was dissolved in methanol (2 mL), then mixed with allyl chloride (300 mg, 4 mmol) as a monomer in methanol (2 mL), and left for a few seconds at room temperature. Then, ethylene glycol

methacrylate (EGDMA) (3.9 g, 20 mmol) was dissolved in methanol (2 mL) (as a cross-linker), and benzoyl peroxide (300 mg) as an initiator) was dissolved in chloroform (2 mL)

and added to the solution to obtain a homogeneous solution, and the mixture was shaken for 5 minutes. Afterwards, N<sub>2</sub> gas was passed for 30 minutes through the mixture to extract oxygen from it. The solution was then placed in a water bath at 60 °C for 6 hours. When the reaction was completed and the Ca-IIP2 was formed as very hard materials, they were left for 24 hours to dry, and then they were crushed and ground by a mortar and pestle. A sieve was used to obtain particles with a diameter of 125-150 µm which were then collected. Calcium ion was extracted from polymers Ca-IIP using a soxhlet of (60:10) (methanol, acetic acid) for weak extraction. After ensuring complete template removal, the polymer was dried for 24 hours at room temperature and collected to be used as a substance in a solid-phase extraction syringe. Each plastic syringe (column) was packed with Ca-MIP (200 mg) and used a 3 mL solution for solid-phase extraction by a peristaltic pump.



**Figure 1:** Steps to prepare the ion imprinted polymeric of  $\text{Ca}^{2+}$

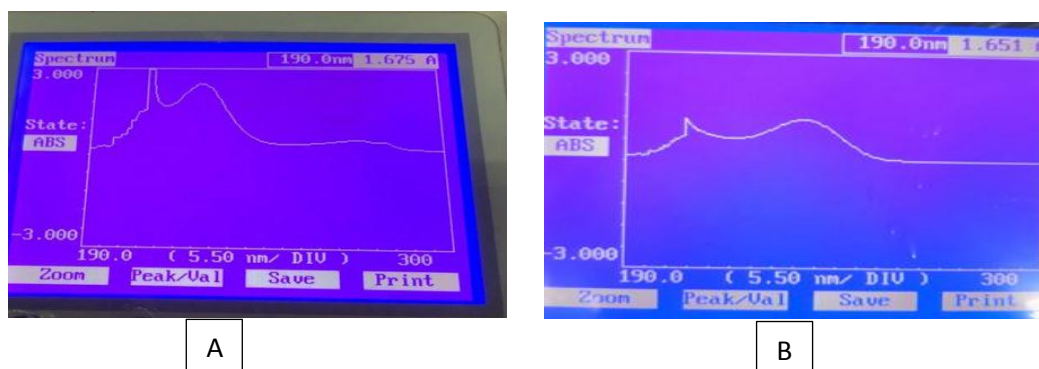
\*For one type of polymeric ionic print, several columns were used A\*

### 2.3. Sampling procedure

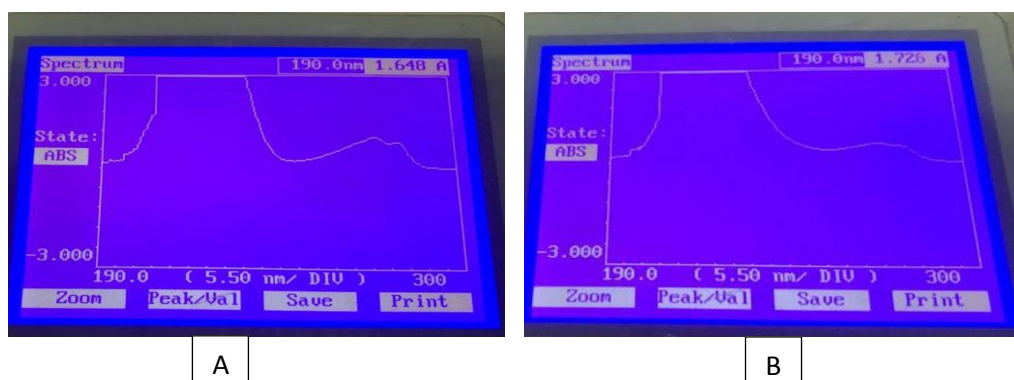
Serial concentrations (20, 40, 60, 80, 100, 120, and 150 ppm) were prepared from  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$  (147 mg, 1 mmol) by dissolving certain weights in methanol in a 100 mL volumetric flask. Calibration curve between the X axis (concentration of  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ ) and the Y axis of its absorption, as shown in Figure 11 and Table 4 using a UV-VIS instrument at 276 nm. Also, three samples were taken before and after treatment (filtered to remove unwanted minutes) (from well water from Hor Rajab, irrigation water from Jadriyah Farms, and Tigris water from the Nahrawan region).

### 3. Results and discussion

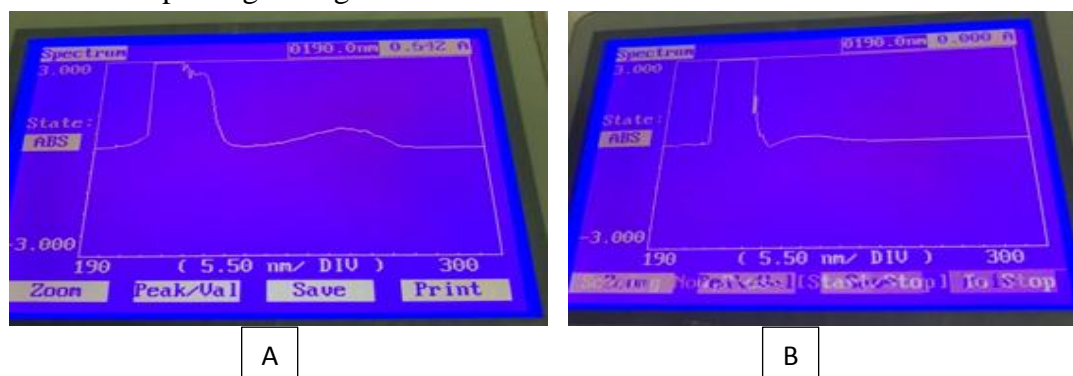
Figures 2, 3, and 4 provide a clear illustration of the benefits of using impressed polymers in SPE for the quantification of the calcium ions after the environmental samples were brought Water from Jadriyah Farms, well water from Hor Rajab, and Tigris water from the Nahrawan region were all passed through a syringe containing Ca-MIP. The residue with the least absorption was measured using a UV-VIS spectrophotometer.



**Figure 2:** A and B, the absorption spectra of calcium ions in irrigation from Jadiriyah Farms water before and after passing through the IIP column



**Figure 3 -** A and B, the absorption spectra of calcium ions in well water from Hor Rajab before and after passing through the IIP column



**Figure 4:** A and B, the absorption spectra of calcium ions in Tigris water from the Nahrawan region before and after passing through the IIP column

### 3.1. FT-IR analysis

To determine the functional groups present in a compound, Fourier Transform Infrared Spectroscopy (FT-IR) is considered an important chemical characterization process. The Ca-MIP FT-IR spectra show several functional groups (before and after removal of calcium ions) in the following Figures. The FT-IR spectra (before and after the elimination of  $\text{Ca}^{+2}$  for IIP) are shown in Figures 5, 6, and 7.

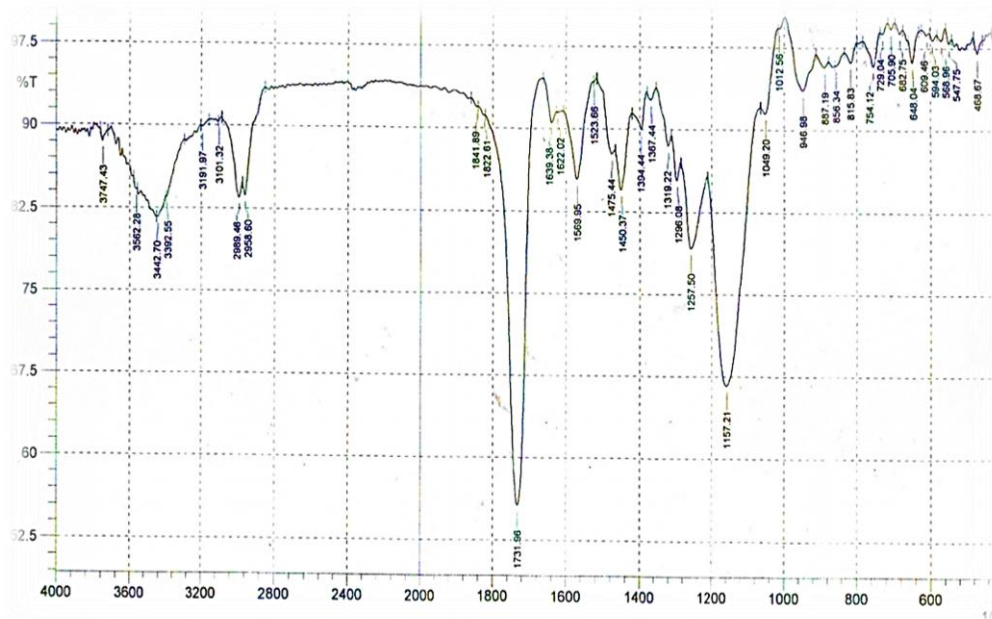


Figure 5: FT-IR spectrum of CaCl<sub>2</sub>·2H<sub>2</sub>O.

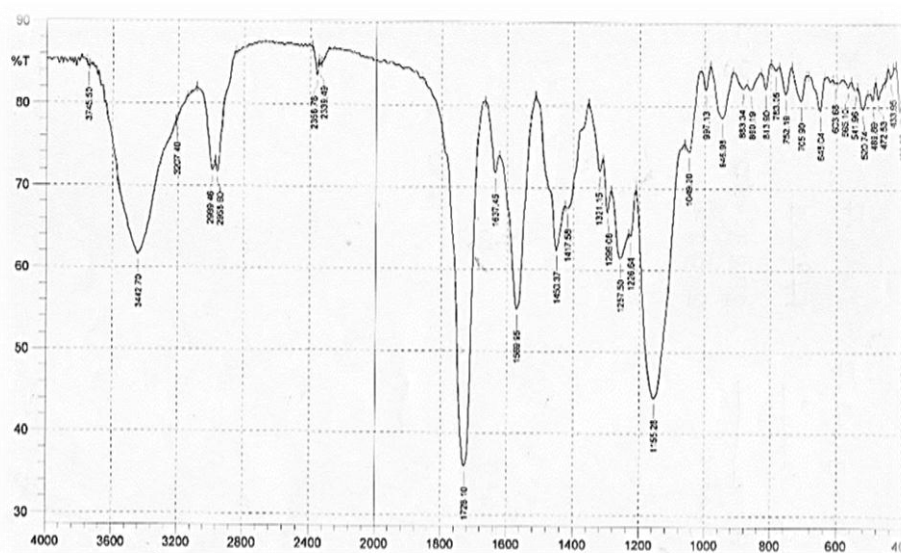


Figure 6: FT-IR spectrum of Ca-IIP1 before removal the ion Ca<sup>2+</sup>

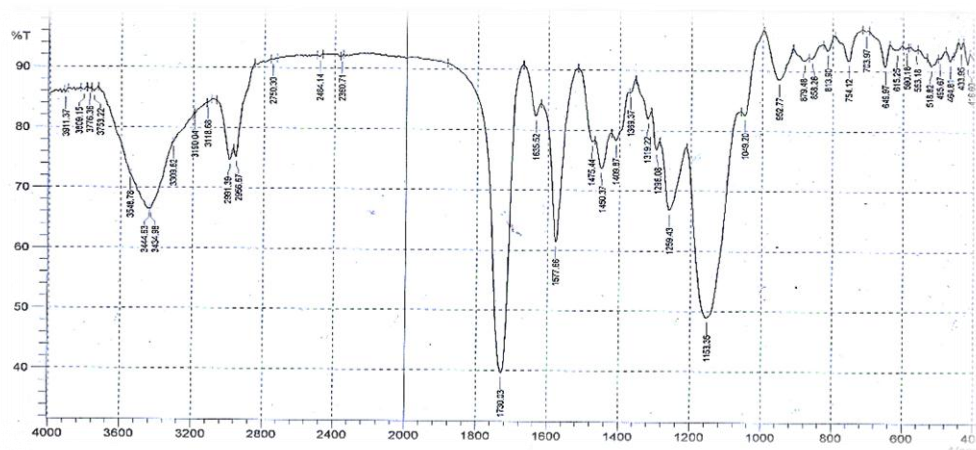


Figure 7: FT-IR spectrum of Ca-IIP before and after extraction (after removal the ion Ca<sup>2+</sup>)

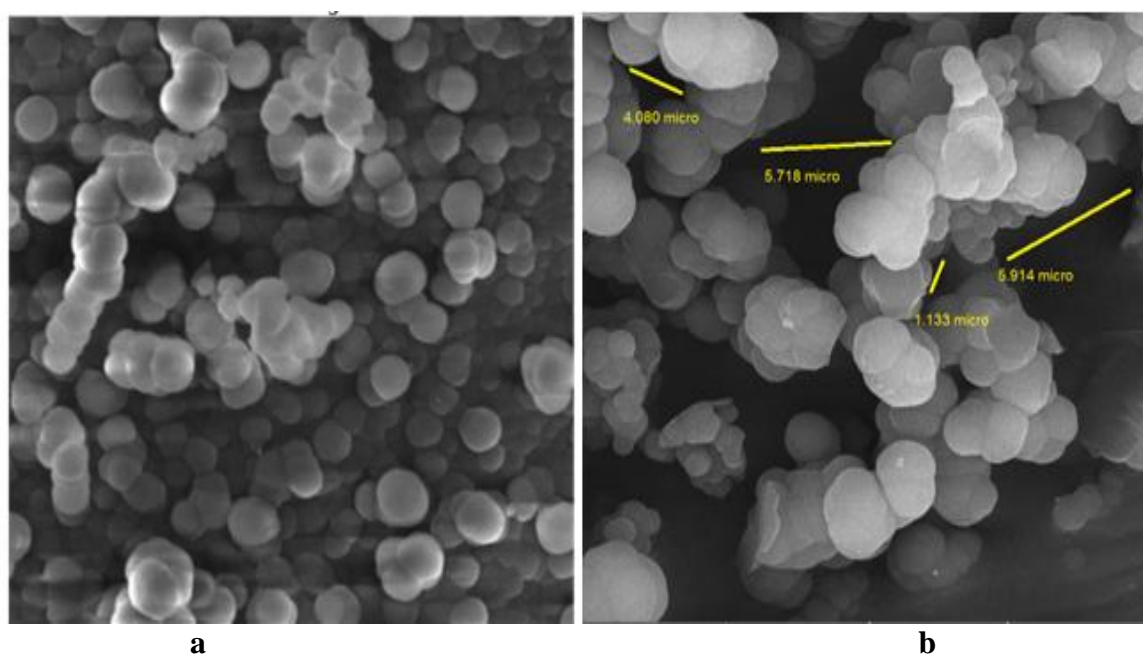
Before calcium removal, a band was visible in the FT-IR spectra at  $1569\text{ cm}^{-1}$  stretching for Ca-Cl,  $3443\text{ cm}^{-1}$  for hydroxyl group stretching,  $1728\text{ cm}^{-1}$  for C=O stretching,  $2989$  and  $2959\text{ cm}^{-1}$  for C-H aliphatic stretching [12]. In comparison, the spectra of the FT-IR after the removal of calcium ions showed the disappearance of the band of Ca-Cl, which indicated the release of the calcium ions and the creation of the molecularly imprinted polymer.

**Table 1:** The most identified peaks of FT-IR spectrum of Ca-IIP using allyl chloride as a functional monomer

Functional Group ( $\nu, \text{cm}^{-1}$ )	$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	Ca-IIP (allyl chloride) before template removal	Ca-IIP (allyl chloride) after template removal
Ca-Cl	1569	1570	-
O-H	3443	3443	3435
C=O	-	1728	1730
C-H	-	2989 2959	2991 2957

### 3.2. Scanning electron microscope (SEM)

A scanning electron microscope (SEM) creates a high-resolution image by scanning the surface of a comparison surface; this figure depicts the morphology of IIP for calcium before and after washing. The figure reveals obvious calcium holes in the sizes eliminated by Soxhlet extraction [13].



**Figure 8:** SEM photograph of the surface of Ca-IIP (allyl chloride), (b) before calcium removal, (a) after calcium removal

**Table 2:** Contrast ratios between salt, monomer, and cross-linker in the preparation of calcium-IIP

		Salt CaCl <sub>2</sub> .2H <sub>2</sub> O	Monomer Allyl chloride	Cross linker EGDMA	Initiator Benzoyl peroxide	Solvent	Result
MIP	%	3.727	16.876	79.395	0.3	6 mL	Pale
	Mmole	0.855	3.871	18.211	0.32	CH <sub>3</sub> OH	white
MIP	%	3.831	16.612	79.556	0.3	6 mL	Pale
	Mmole	0.912	3.954	18.936	0.32	CH <sub>3</sub> OH	white
MIP	%	4.048	16.211	83.45	0.3	6 mL	Pale
	Mmole	0.999	3.99	19.675	0.32	CH <sub>3</sub> OH	white
NIP	%	-	16.211	83.45	0.3	6 mL	Pale
			4	19.675	0.32	CH <sub>3</sub> OH	white

\*The optimum ratios employed in the synthesis of Ca-ion-imprinted polymers (IIPs) and non-imprinted polymers (NIPs) are summarized in Table2. After the calcium ion is removed, the control NIPs and IIPs, however, exhibit the same spectra and structural similarities. This demonstrates that removing the template molecule and leaving particular recognition binding sites in the polymer structure may be accomplished by washing the IIP particles in a (methanol:acetic acid, 60:10) solution using the Soxhlet extraction method.

Q: Capacity

Q free: Capacity at final concentration

$$Q = [(C_i - C_f) V_s] / W_{mip}$$

C<sub>i</sub> = Initial concentration of CaCl<sub>2</sub>.2H<sub>2</sub>O (μmole/mL)

C<sub>f</sub> = Final concentration of CaCl<sub>2</sub>.2H<sub>2</sub>O (μmole/mL)

V<sub>s</sub> = Volume of solution tested (mL)

$$Q / C_f = (Q_{max} - Q) / K_d$$

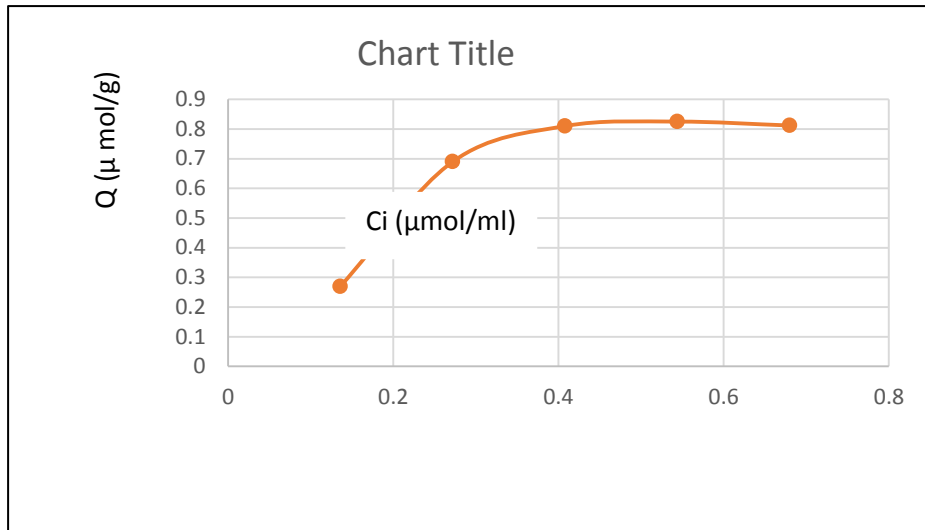
Q<sub>max</sub> = Maximum capacity

K<sub>d</sub> = Dissociation constant at binding side

**Table 3:** The optimal synthesis conditions for the ionic imprinted polymer for Ca-IIP (allyl chloride) developed in this study using UV-VIS instruments

Ca-IIP (allyl chloride)					
Mass of MIP (mg)	C <sub>i</sub> (ppm)	C <sub>i</sub> (μM)	C <sub>free</sub> (μM)	Q (μMole/g)	Q <sub>free</sub> (mL/g)
0.2	20	0.136	0.127	0.270	2.125
	40	0.272	0.249	0.690	2.771
	60	0.408	0.354	0.810	2.288
	80	0.544	0.4890	0.825	1.687
	100	0.680	0.626	0.812	1.293





**Figure 9:** The relation between initial concentration  $C_i$  ( $\mu\text{mol/mL}$ ) and capacity  $Q$  ( $\mu\text{mol/g}$ )

$$1/q_e = 1/Q_o + 1/Q_o K_L C_e \quad [14]$$

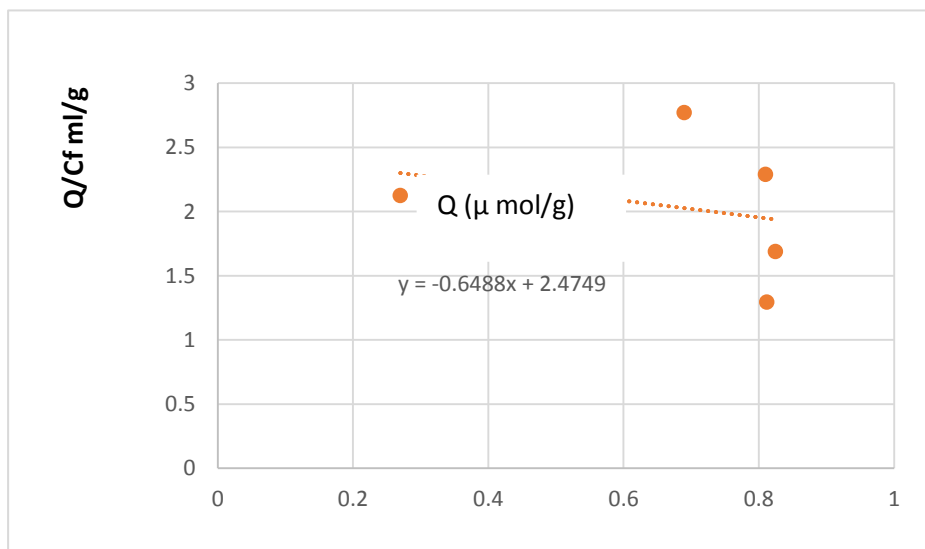
$C_e$  = The equilibrium concentration of adsorbate ( $\text{mg/L}^{-1}$ ).

$q_e$  = The amount of metal adsorbed per gram of the adsorbent equilibrium ( $\text{mg/g}$ ).

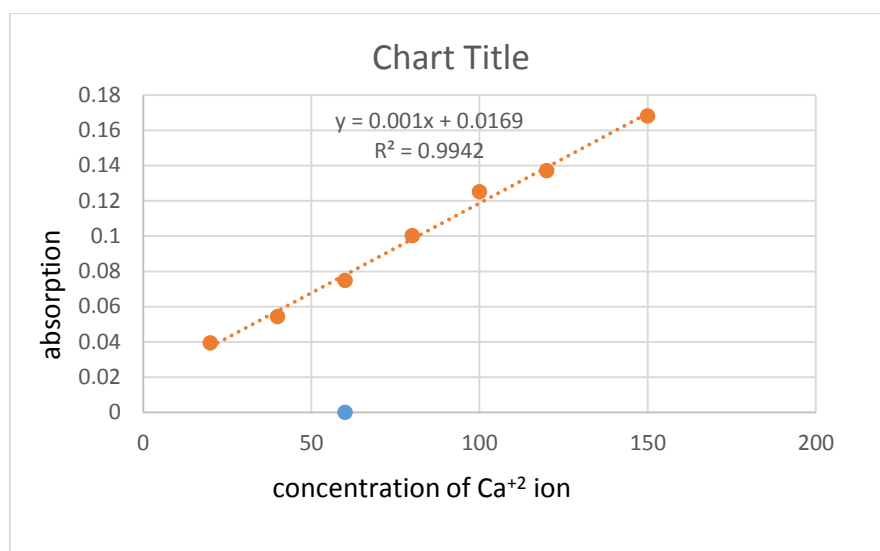
$Q_o$  = Maximum monolayer coverage capacity ( $\text{mg/g}$ ).

Where  $K_L$  = Langmuir isotherm constant ( $\text{L/mg}$ ).

The Langmuir equation was chosen because the findings demonstrated that the concentration rises proportionately to the rise in absorbance, yielding one slope.



**Figure 10:** The relation between capacity  $Q$  ( $\mu\text{mol/g}$ ) and  $Q/C_f$  ( $\text{mL/g}$ )



**Figure 11:** Calibration curve between concentrations of calcium ions at standard ppm and their absorption in a UV- VIS spectrophotometer

$$\text{Slop} = -1/kd$$

$$-0.6488 = -1/ kd = 1.5413$$

$$\text{Intercept} = Q_{\max}/kd$$

$$2.4749 = Q_{\max}/1.5413$$

$$Q_{\max} = 3.8145 \mu\text{mol/g}$$

**Table 4:** The relation between concentration of  $\text{Ca}^{2+}$  ion ppm and absorption

Concentration of $\text{Ca}^{+2}$ ion ppm	Absorption
20	0.0394
40	0.0543
60	0.0748
80	0.1002
100	0.125
120	0.137
150	0.168

\*Experiment with time and temperature did not work due to

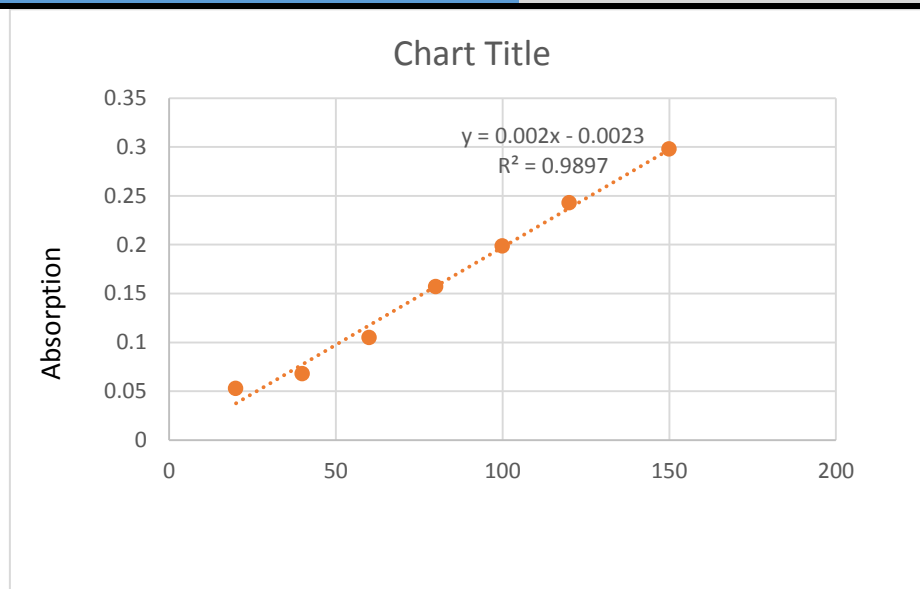
$$Q = [(C_i - C_f) V_s] / W_{\text{mip}}$$

### 3.3. Atomic absorption spectroscopy (AAS)

Standard solutions with concentrations of 20, 40, 60, 80, 100, 120, and 150 ppm were prepared and measured by atomic absorption at wavelength  $\lambda$  422.7 nm, as shown in Figure 12 and Table 5.

**Table 5 :** The relation between concentration of Ca<sup>+2</sup> ion ppm and absorption

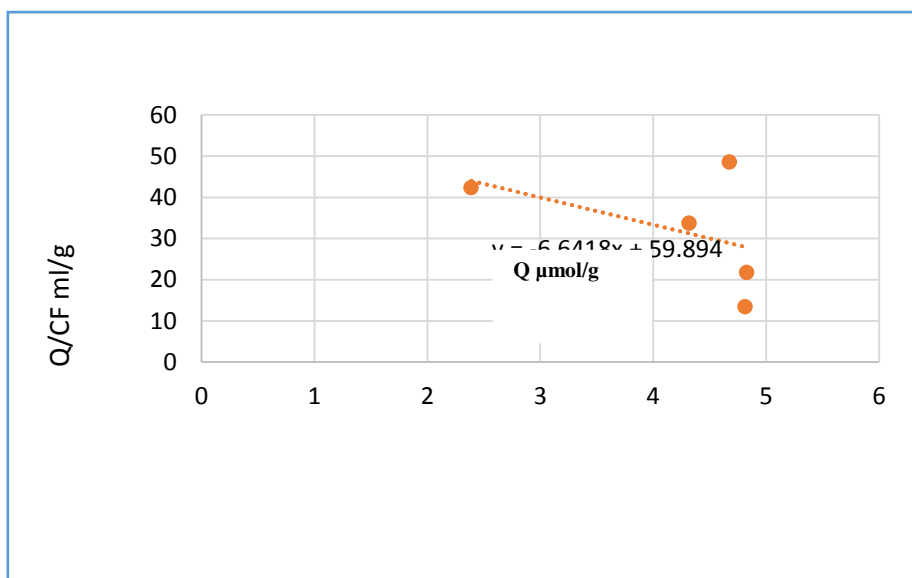
Concentration of Ca <sup>+2</sup> ion ppm	Absorption
20	0.053
40	0.068
60	0.105
80	0.157
100	0.198
120	0.243
150	0.298



**Figure 12:** Calibration curve between concentrations of calcium ions at standard ppm and their absorption in a UV- VIS spectrophotometer

**Table 6:** The optimal synthesis conditions for the ionic imprinted polymer for calcium developed in this study used AAS

Mass of MIP (mg)	IIP - Ca (Allyl chloride)				
	C <sub>i</sub> (ppm)	C <sub>i</sub> (μM)	C <sub>free</sub> (μM)	Q (μMole/g)	Q <sub>free</sub> (mL/g)
0.2	20	0.136	0.0564	2.388	42.340
	40	0.272	0.128	4.320	33.750
	60	0.408	0.096	4.675	48.551
	80	0.544	0.222	4.830	21.756
	100	0.680	0.359	4.815	13.412



**Figure 13:** The relation between capacity Q (μmol/g) and Q/Cf (mL/g)

Slope = -1/kd

-6.6418 = -1/ kd

Kd = 1/ 6.6418

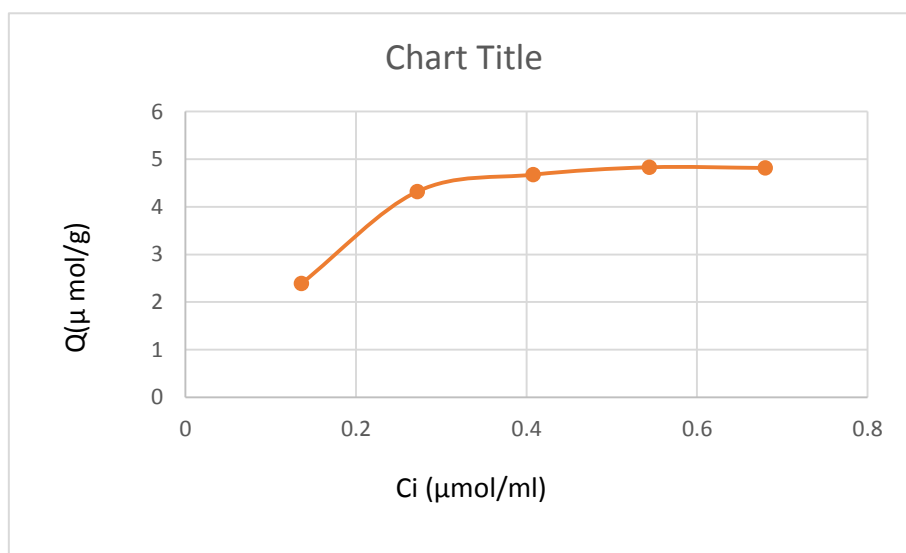
Kd = 0.15056

Intercept = 59.894

Intercept = Q<sub>max</sub>/ kd

Q<sub>max</sub> = 59.894×0.15056

Q<sub>max</sub> = 9.01773μmol/g



**Figure 14:** The relation between initial concentration Ci (μmol/mL) and capacity Q (μmol/g)

**Table 7:** The UV-VIS measurement results for a water sample

Water source	Absorption (mean)	RSD (%) = $(\delta n-1/\text{Mean}) * 10$ precision	Rec (%) = (practical value/True value)*100 accuracy	Re% = 100-Rec
water well	0.0390	0.256	98.9	1.1
Irrigation water	0.0529	0.189	97.4	2.6
Tigris water (Nahrawan region)	0.0720	0.138	96.2	3.8

**Table 8:** Measurement results for the water sample used A.A.S

Water source	Absorption (mean)	RSD (%) = $(\delta n-1/\text{Mean}) * 100$ precision	Rec (%) = practical value/True value)*100 Accuracy	Re (%) = 100-Rec
water well	0.108	0.925	102.85	-2.85
Irrigation water	0.0559	0.178	105.47	-5.47
Tigris water (Nahrawan region)	0.0651	0.153	95.73	4.27

**Table 9:** Compare the capacity of two analytical techniques, atomic absorption and IIP determination of  $\text{Ca}^{+2}$  ions in water sources

Water source	Capacity Q $\mu\text{mol/g}$ for atomic technique	Capacity Q $\mu\text{mol/g}$ for UV-Visible
Water well	0.450	0.360
Irrigation water	0.619	0.615
Tigris water (Nahrawan region)	0.495	0.450

\*

According to the above table, we note that the capacitance values in the atomic absorption technique are greater than the capacitance values in the UV-VIS technique. This is because of the sensitivity of the device, and it has a lower sensitivity limit than the UV-VIS technique, but in general, the results are acceptable and close to each other. This is evidence of the method's suitability for use in assessing and quantifying calcium ions.

#### 4. Conclusion

Bulk polymerization was used to create a novel calcium-IIP. EGDMA was chosen as the cross-linker and allyl chloride as the functional monomer. In addition, benzyl peroxide was utilized as an initiator when chloroform was the solvent. The ideal calcium (II) ion to monomer and cross-linker dosage molar ratios was investigated. The three-dimensional network structure of polymers and their unpredictable shapes were studied using SEM. The results of FT-IR demonstrated that the Ca (II) ion was successfully eluted by a solution of methanol and acetic acid (60:10 v/v). The exceptional stability and regeneration capabilities of calcium-IIP are illustrated by the fact that the elution process has little to no impact on the chemical characteristics of the polymer or the shape of the cavity.

#### References

- [1] M. Jaishankar, T. Tseten, N. Anbalagan, B. B. Mathew, and K. N. Beeregowda, "Toxicity, mechanism and health effects of some heavy metals," *Interdisciplinary toxicology*, vol. 7, no. 2, pp. 60-72, 2014.
- [2] J. A. Danner, "Predictive model to estimate ionized calcium from routine serum biochemical profiles in dogs," 2017.

- [3] U. Trebbe, M. Niggemann, K. Cammann, G. C. Fiaccabrino, M. Koudelka-Hep, S. Dzyadevich, O. Shulga, "A new calcium-sensor based on ion-selective conductometric microsensors-membranes and features," *Fresenius' Journal of Analytical Chemistry*, vol. 371, no. 6, pp. 734-739, 2001.
- [4] Y. K. Al-Bayati, "Preparation of Selective Sensors for Cyproheptadine Hydrochloride based on Molecularly Imprinted Polymer used N, N-Diethylaminoethyl Methacrylate as Functional Monomer," *Eurasian Journal of Analytical Chemistry*, vol. 13, no. 5, pp. 1-10, 2018.
- [5] M. Andaç, R. Say, and A. Denizli, "Molecular recognition based cadmium removal from human plasma," *Journal of Chromatography B*, vol. 811, no. 2, pp. 119-126, 2004.
- [6] A. J. Al-Safi and Y. K. Al-Bayati, "Synthesis and Characterization of molecularly imprinted Polymer for Tramadol HCL Using Acryl Amide And 2-Hydroxyethyl Meth Acrylate AS Monomers," *Current Issues in Pharmacy and Medical Sciences*, vol. 31, no. 2, pp. 81-88, 2018.
- [7] S. Abd Jaber and Y. K. Al-Bayati, "Synthesis of the New Naproxen Selective Electrode Based on Imprinted Polymer Using Different Monomers and its Determination at Pharmaceutical Preparation," *Iraqi journal of market research and consumer protection*, vol. 12, no. 2, PP. 1-11, 2020.
- [8] A. Al Fatease *et al.*, "Label-free electrochemical sensor based on manganese doped titanium dioxide nanoparticles for myoglobin detection: biomarker for acute myocardial infarction," *Molecules*, vol. 26, no. 14, p. 4252, 2021.
- [9] S. Ismaeel and Y. K. Al-Bayati, "Determination of Trace Metformin in Pharmaceutical Preparation Using Molecularly Imprinted Polymer based Pvc-Membrane," *Eurasian Chemical communications Journal*, vol.3, no. 11, p. 812-830, 2021.
- [10] V. Camel, "Solid phase extraction of trace elements," *Spectrochimica Acta Part B: Atomic Spectroscopy*, vol. 58, no.10, pp. 1177-1233, 2003.
- [11] M. F. Alam, Z. A. Begum, Y. Furusho, H. Hasegawa, and I. M. Rahman, "Selective separation of radionuclides from environmental matrices using proprietary solid-phase extraction systems: A review," *Microchemical Journal*, vol.181, no.10, p. 107637, 2022.
- [12] D. Agarwal and V. K. Kasana, "Synthesis and FT-IR, SEM, EDS Studies of Heterogeneous Catalyst-CaCl<sub>2</sub>. 2H<sub>2</sub>O Supported on Rice Husk: A Highly Efficient and Economical Catalyst for N-Formylation of Amines at Room Temperature," *International Research Journal of Pure and Applied Chemistry*, vol. 18, no. 1, pp. 1-10, 2019.
- [13] E. Zaheer, S. Hassan, H. Shareef, A. Naz, A. Hassan, and K. Qadeer, "Scanning electron microscopy (SEM) and atomic absorption spectroscopic evaluation of Raphanus sativus L. seeds grown in Pakistan," *Pakistan Journal of Pharmaceutical Sciences*, vol. 34, no. 2, pp. 545-552, 2021.
- [14] A. Dada, A. Olalekan, A. Olatunya, and O. Dada, "Langmuir, Freundlich, Temkin and Dubinin-Radushkevich isotherms studies of equilibrium sorption of Zn<sup>2+</sup> unto phosphoric acid modified rice husk," *IOSR Journal of applied chemistry*, vol. 3, no. 1, pp. 38-45, 2012.