

Rapid determination of magnesium and calcium ions in saline and seawater

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ABSTRACT

Access to fresh water from seawater treatment is crucial for sustaining human life. Furthermore, Ca and Mg in seawater represent valuable resources for agricultural and industrial purposes, warranting thorough analysis and recovery efforts. This study introduces the use of ortho-cresolphthalein complex one (o-CPC) for the rapid determination of Ca and Mg concentrations in saline and seawater samples. This approach utilizes a basic complexation reaction and follows the concept of a swift, point-of-care testing kit used to analyze samples, specifically biological specimens. The methodology was meticulously examined and optimized, encompassing key parameters such as reagent concentration, employment of masking agents, determination of optimal wavelengths, and assessments of accuracy and precision. The resulting standard curve exhibited high linearity ($R^2 > 0.995$) across 0.5–5 mg. L⁻¹ for Ca and 0.5–10 mg. L⁻¹ for Mg. Notably, the method demonstrated good repeatability (% RSD < 5%) and achieved high recovery rates (95%–105%) when tested on spiked samples within real sample matrices. A comparative analysis between the o-CPC method and the F-AAS method (in accordance with the SMEWW 3111B:2017 standard) was conducted using three seawater samples, revealing no significant discrepancies in the analysis of Mg and Ca with a confidence level of 95%.

Key words: magnesium, calcium, o-CPC method, seawater

1 INTRODUCTION

The escalating issue of salinity presents a critical challenge that directly impacts the livelihoods of coastal communities, exacerbating freshwater scarcity. With its extensive 3,000 km coastline, Vietnam is directly affected by this challenge, notably in the Mekong Delta region, which is characterized by an intricate network of rivers bordering the sea. The dry season intensifies the complexity of saltwater intrusion, exacerbated by reduced upstream water flows, less rainfall (under 10 mm/month), and tidal inflows carrying seawater inland. Salinity levels surpass 45% along the Tien and Hau Rivers, peaking at 70% near river mouths and reaching 85% intrusion in certain areas. Consequently, ensuring a sustainable freshwater supply for irrigation and domestic usage requires ongoing research and development of desalination technologies¹.

Seawater predominantly comprises approximately 99.2% of ions, notably sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), chlorine (Cl), and sulfate (SO₄) ions. Na and Cl ions, which are the most abundant, are extensively utilized in saltwater treatment processes. Furthermore, brine contains economically valuable materials such as calcium carbonate and magnesium sulfate, making it viable for industrial and agricultural applications^{2,3}.

Calcium (Ca) and magnesium (Mg) are also essential trace elements present in seawater and exert a pivotal influence on establishing and regulating metabolic cycles in marine ecosystems. Additionally, the examination of magnesium and calcium provides a fundamental framework for evaluating diverse pertinent factors, particularly focusing on the desalination capabilities of various technologies. Consequently, there is a critical need for techniques to accurately determine the concentrations of these ions, especially in seawater or saline environments.

Various methodologies have been devised to evaluate the efficacy of desalination technologies by assessing the concentrations of Ca and Mg in seawater pre- and posttreatment. The titration method, outlined in the Vietnamese standard TCVN 6,201:1995 (ISO 6,059:1984), employs EDTA as a reagent with Erichrome Black T as an indicator. This method is widely utilized for the concentration analysis of these two ions in water owing to its simplicity. Nonetheless, the titration method suffers from limitations in accurately quantifying ions and is susceptible to interference from other metals that can form complexes with EDTA. As a result, measuring the levels of Ca

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and Mg in high-salinity water, which contains complex compositions of cations, is challenging. Alternatively, according to TCVN 6,201:1995, atomic absorption spectroscopy is commonly referenced, often involving the incorporation of lanthanum to eliminate interferences in specific sample matrices. Overall, this approach demonstrates a strong ability to selectively and effectively analyze the presence of Mg and Ca in seawater. Nevertheless, the abundance of salts in seawater can lead to the accumulation of salt deposits on the atomic absorption component, impeding the progress of the analysis procedure. Additionally, ion chromatography, specified in TCVN 6,660:2000 (ISO 14,911:1998), is utilized for analyzing Ca and Mg in brine solutions. Some drawbacks of this method include a restricted operational range, vulnerability to organic compound interference, and reliance on the lifespan of the chromatographic column. In pursuit of rapid and straightforward determination, this study adopts molecular absorption spectroscopy as the primary analytical method for assessing the Mg and Ca contents in seawater. The findings are juxtaposed with those obtained through alternative techniques.

Calcium and magnesium ions readily form complexes with various reagents, such as methymolblue, cagmagite, EDTA, and ortho-cresolphthalein complexone (o-CPC)⁴. Among these methods, the colorimetric method employing o-CPC as a reagent is widely favored for quantifying Ca and Mg contents, especially in biological test kits commonly utilized in medical diagnostics, as corroborated by previous studies⁵. In this study, o-CPC (H_2Z^{4-}) was chosen owing to its rapid and straightforward color development process, coupled with the stability of the resulting complex products, aligning with the objectives outlined in the study.

MATERIALS AND METHODS

Materials and instruments

Instrumentation

Absorbance measurements were conducted using a dual-beam spectrophotometer (Shimadzu Model 1800s) equipped with a 1-cm cuvette alongside a Perkin Elmer AAnalyst 700 flame atomic spectrophotometer.

Materials

Diethanolamine (>99%) was purchased from Xilong Scientific, China, and diethylamine (99%) from Scharlau, Spain, served as the buffer solvent. Stock standard solutions of Ca^{2+} (1,000 mg. L^{-1}) and Mg^{2+} (1,000 mg. L^{-1}) were supplied by Merck, Germany. New bottles containing these analytes were

stored in a desiccator before use. o-CPC reagent (Fluka, USA) supplemented with 8-hydroxyquinoline (>99.5%) was chosen as the masking agent and was obtained from Shanghai Zhanyun, China. Distilled water was used throughout all the experimental procedures.

Reagent preparation

Preparation of the working standard solution involved diluting stock solutions of Ca^{2+} and Mg^{2+} , each at a concentration of 1,000 mg. L^{-1} , is used to obtain solutions at 50 mg. L^{-1} in distilled water.

To prepare 0.20 mM ortho-cresolphthalein complexone, 32.5 g of ortho-cresolphthalein complexone was dissolved in 250 mL of buffer solution at pH 11.5. This buffer solution was prepared using diethylamine at a concentration of 24 mM and diethanolamine at 0.5 M, with a ratio of diethylamine:diethanolamine:water = 1:20:400 (v/v).

To prepare 0.10 M 8-hydroxyquinoline, 750 mg of 8-hydroxyquinoline and 0.5 mL of sulfuric acid were dissolved in 50 mL of distilled water. The resulting solution was stored at 4°C in a dark vessel.

Sample preparation

Seawater samples were collected from three different locations in accordance with standards TCVN 6663-1:2011, TCVN 6663-3:2016, and TCVN 5,998:1995. These samples were stored under refrigeration at temperatures ranging from 1 to 5°C.

Subsequently, the samples underwent filtration and dilution with distilled water, employing an appropriate dilution factor.

General procedure

To determine the total concentration of Ca^{2+} and Mg^{2+} , 2 mL of the sample was added to a volumetric flask, followed by the addition of 3.5 mL of 0.2 mM o-CPC reagent to initiate the colorimetric reaction.

For Ca^{2+} determination alone, 2 mL of the sample was added to a volumetric flask with 1 mL of 0.2 mM o-CPC reagent and 0.2 mL of 0.1 M 8-hydroxyquinoline masking agent to facilitate color development.

Upon the formation of the Ca-Mg-o-CPC complex, the absorbance of the solution was measured at the wavelength of maximum absorption ($\lambda_{max} = 572$ nm).

Optimization experiment

Investigate wavelength

Under optimized conditions, the maximum absorption wavelengths for both the Ca-OCPC and Mg-OCPC complexes were determined by conducting a colorimetric reaction alongside an equivalent reagent blank sample across the 400–700 nm wavelength range. Sample concentrations of 3.00 mg. L⁻¹ for calcium Ca²⁺ and 4.00 mg. L⁻¹ for magnesium Mg²⁺ were employed in this investigation.

Calibration curve

The calibration curve for the individual quantification of calcium and magnesium Ca²⁺ and Mg²⁺ ions was developed within concentration ranges of 0.5–5 and 0.5–10 mg, respectively. L⁻¹, respectively. The experiments were conducted in triplicate.

Investigate reagent concentration

The experiments were performed in a buffer environment with a pH of 11.5, focusing on the color reaction between Ca²⁺, Mg²⁺ and o-CPC at various o-CPC concentrations ranging from 5 to 150 mg.L⁻¹. The change in the absorbance of the resulting complex was measured through optical measurements at λ_{max} . Each cation was assessed at two concentration levels: 5 and 10 mg. L⁻¹.

Investigate the 8-hydroxyquinoline concentration

This assessment was conducted concurrently using a sample containing 2.00 mg. L⁻¹ Ca²⁺ and a laboratory sample of a mixture with a Ca²⁺ concentration maintained at 2.00 mg. L⁻¹ and a Mg²⁺ concentration of 6.00 mg. L⁻¹, with a Ca:Mg ratio of 1:3, representing the brine sample. The concentrations of 8-hydroxyquinoline investigated were prepared by weighing the appropriate amounts of reagent (ranging from 12.50 to 500.00 mg), dissolving them in 0.25 mL of concentrated H₂SO₄, and subsequently diluting them to 25.00 mL with distilled water.

Investigate the effectiveness of 8-hydroxyquinoline in removing magnesium interference at different Mg:Ca ratios

The research sample solution was created by maintaining a constant volume of 1 mL of 100 mg. L⁻¹ Ca²⁺ solution while varying the volume of 100 mg. L⁻¹ Mg²⁺ solution within the range of 1–8 mL. The colorization process was performed following a methodology similar to that outlined in section 2.4, aimed at assessing the efficacy of the 8-hydroxyquinoline masking agent in eliminating interference.

Method validation

The Con Dao seawater sample was selected to assess the repeatability of this method. The experiment was performed six times over three days. A 3% NaCl solution served as the blank. Standard solutions containing Ca²⁺ and Mg²⁺ at concentrations of 200 and 400

mg. L⁻¹, respectively, were added. The accuracy of the method was evaluated based on the recovery rate.

Application of the o-CPC method for determining Ca and Mg in seawater

The Ca²⁺ and Mg²⁺ contents in seawater samples were analyzed using the photometric method with the o-CPC coloring reagent and the comparative method F-AAS (following the SMEWW 3111B:2017 standard). A comparative assessment was conducted to analyze the outcomes of both approaches.

RESULTS

Wavelength

The UV-VIS absorption spectra of complex samples, including Ca-OCPC at 3.00 mg. L⁻¹ and Mg-OCPC at 4.00 mg. L⁻¹, alongside their respective blank reagent samples, are depicted in Figure 1. Overlapping absorption peaks in the 500–600 nm region were observed.

Calibration curve

The repeatability and linearity were good, with an RSD <5% and a high correlation coefficient (R² = 0.9989 and R² = 0.9997) (Figure 2 and Figure 3).

Effect of the reagent concentration

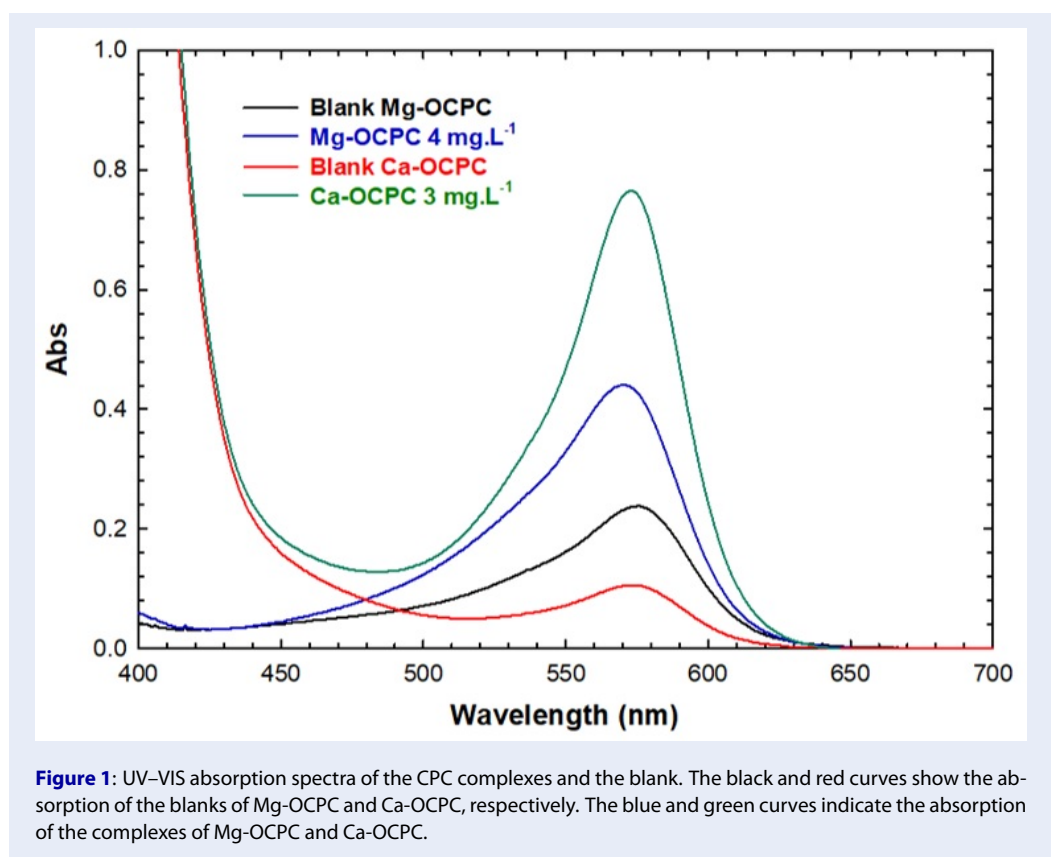
The optical density of the solution was influenced by the reagent concentration. Figure 4 shows that as the reagent volume remains fixed, increasing its concentration leads to a corresponding increase in the optical absorption of the Mg-OCPC and Ca-OCPC complexes. Depending on the sample's Ca²⁺ and Mg²⁺ concentrations, the absorbance trends vary with the reagent concentration.

For Mg²⁺, at concentrations of 5 and 10 mg. L⁻¹, no discernible difference was observed when reagent volumes were added at concentrations ranging from 5 to 50 mg.L⁻¹. An optical density of 5 mg. L⁻¹ Mg solution remained relatively constant, while that of the 10 mg. L⁻¹ Mg solution exhibited increased optical density with increasing reagent concentration, up to 75 mg. L⁻¹ o-CPC.

A similar upward trend in absorbance was noted for Ca²⁺, with the critical reagent concentration for Ca being 5 mg. L⁻¹ and Ca 10 mg. L⁻¹ was determined to be 75 and 100 mg. L⁻¹, respectively.

Effect of the 8-hydroxyquinoline concentration

In this study, the influence of the 8-hydroxyquinoline masking agent concentration was examined. The data presented in Figure 5 indicate that in the absence of the masking agent, the optical absorbance of the sample solution increases. However, upon the addition of 8-hydroxyquinoline, the absorbance of the sample



254 solutions gradually decreased. For this study, a rec-
 255 ommended reagent concentration of 15,000 mg. L⁻¹
 256 was identified.

257 *Effectiveness of 8-hydroxyquinoline in removing mag-*
 258 *nesium interference*

259 The absorption of the solution was affected by the
 260 Mg:Ca ratio. As shown in Figure 6, the solu-
 261 tion absorbance slightly decreased, while the opti-
 262 cal density of the complex remained stable within
 263 the Mg:Ca ratio range of 1:1–3:1. A higher Mg:Ca
 264 ratio corresponded to reduced effectiveness of 8-
 265 hydroxyquinoline, leading to instability in the solu-
 266 tion's hue and rendering quantification unfeasible.
 267 Therefore, a recommended Mg:Ca ratio of 1:3 was
 268 identified.

269 *Precision and accuracy*

270 This method has good repeatability and %RSD values
 271 following AOAC regulations (Table 1).

272 The recovery was within the specified range of the
 273 AOAC (95–105%) (Table 2).

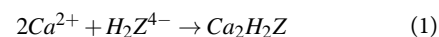
274 *Qualify Ca and Mg in seawater*

275 The analysis of seawater samples collected from
 276 three different sea areas, conducted using the o-CPC
 277 method, revealed consistent fluctuations in the Mg:Ca

ratio, ranging between 3:1 and 4:1. This observed ra-
 278 tio aligns with established publications regarding ion
 279 concentrations in seawater. Specifically, the Ca²⁺
 280 concentrations in the seawater samples varied be-
 281 tween 263 and 344 mg. L⁻¹, while the Mg²⁺ con-
 282 centrations ranged from 1,014 to 1,345 mg.L⁻¹. For
 283 the comparative method, Ca²⁺ and Mg²⁺ were ana-
 284 lyzed by measuring the absorbance of the atom vapor
 285 at wavelengths of 422.7 and 285.2 nm, respectively, in
 286 the presence of La. The findings are presented in Ta-
 287 ble 3.
 288

289 **DISCUSSION**

290 The results of the reagent investigation align with
 291 those of Cathryn M. Corns' theory, which is based
 292 on the interaction between Ca²⁺, Mg²⁺ and o-CPC⁶.
 293 Notably, the reaction forms Mg-OCPC and Ca-
 294 OCPC in a molar ratio of 2:1, as depicted in Eqs. 1
 295 and 2, showcasing the reaction processes.



296 The concentration of o-CPC, which ranged from 125
 297 to 1,500 mg L⁻¹, was stable, and the absorbance of the

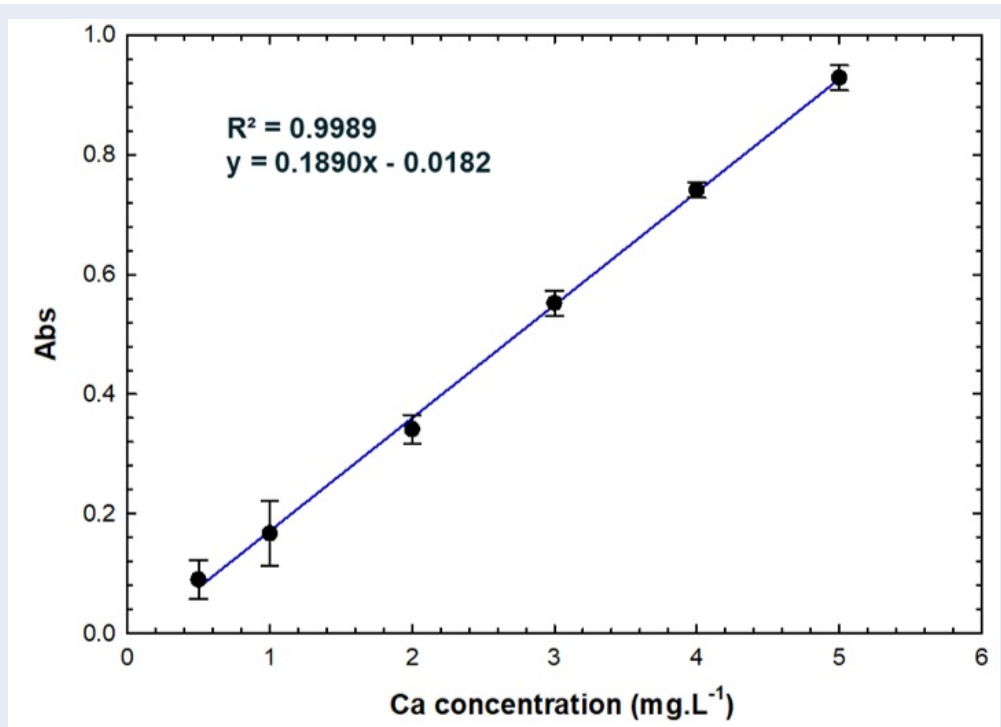


Figure 2: Calibration curve for Ca using the o-CPC method with a concentration range of 0.5 – 5 mg. L⁻¹. The error bar is standard deviations.

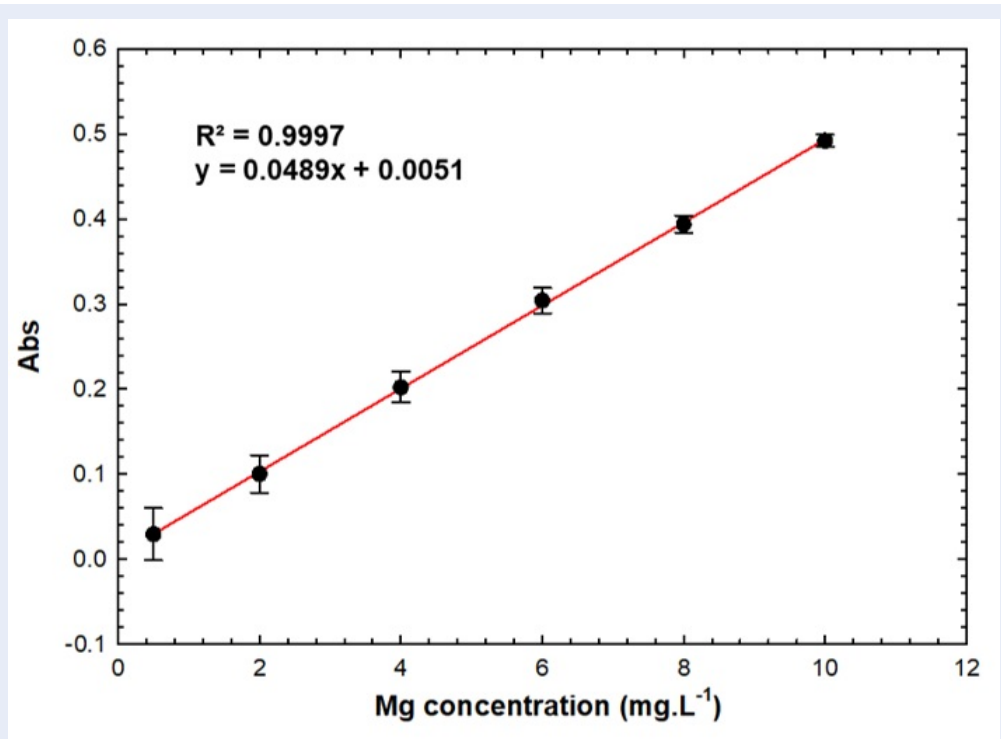


Figure 3: Calibration curve for Mg using the o-CPC method with a concentration range of 0.5 – 10 mg. L⁻¹. The error bar is standard deviations.

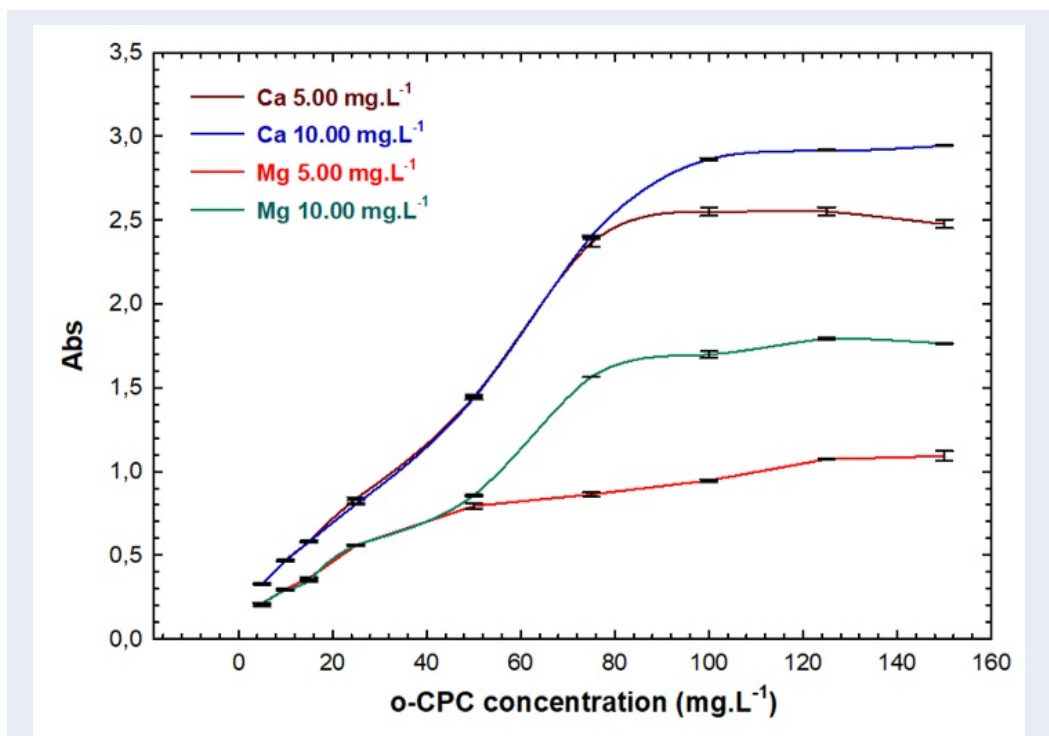


Figure 4: Effect of the o-CPC reagent concentration on cation concentrations. The respective changes in absorption of cation Ca^{2+} are illustrated by the brown and blue curves, representing two distinct concentrations (5 mg.L^{-1} and 10 mg.L^{-1}). The red and green curves get a similar expression for Mg at concentrations of 5 mg.L^{-1} and 10 mg.L^{-1} . The error bar is standard deviations.

Table 1: Results of the Con Dao seawater sample analyzed over 3 days

Cation	Ca		Mg	
Day	C ± SD	%RSD	C ± SD	%RSD
Day 1 (n = 6)	330 ± 13	3.9	1,300 ± 29	2.2
Day 2 (n = 6)	311 ± 16	5.1	1,291 ± 43	3.3
Day 3 (n = 6)	296 ± 14	4.7	1,248 ± 28	2.2

Table 2: Recovery results

Recovery	Average	%RSD
H% Ca	102.0	3.2
H% Mg	100.3	3.0

298 Mg-OCPC and Ca-OCPC complexes remained constant. Within this range, Ca^{2+} and Mg^{2+} in the solution fully reacted to form complexes with the reagent. 300 Based on this observation, 130 mg L^{-1} was identified as the optimal reagent concentration (Figure 4). 302

303 8-Hydroxyquinoline (8-HQ) was chosen as a preferred masking agent for Ca^{2+} quantification alongside o-CPC owing to its diminished ability to form 305 chelated complexes with Ca^{2+} compared to o-CPC. 306

307 Conversely, Mg^{2+} exhibited a stronger chelating affinity for 8-HQ than for o-CPC (Figure 5). 308

309 The Mg:Ca ratio plays a crucial role in the color development process and the efficacy of 310 8-hydroxyquinoline in eliminating interference. 311 Elevated ratios beyond 3:1 generated significant amounts of Mg-8HQ and released H^+ , rendering 312 the Ca-OCPC complex unstable⁷. Additionally, 313 competition between the ligands o-CPC and 8-HQ 314 315

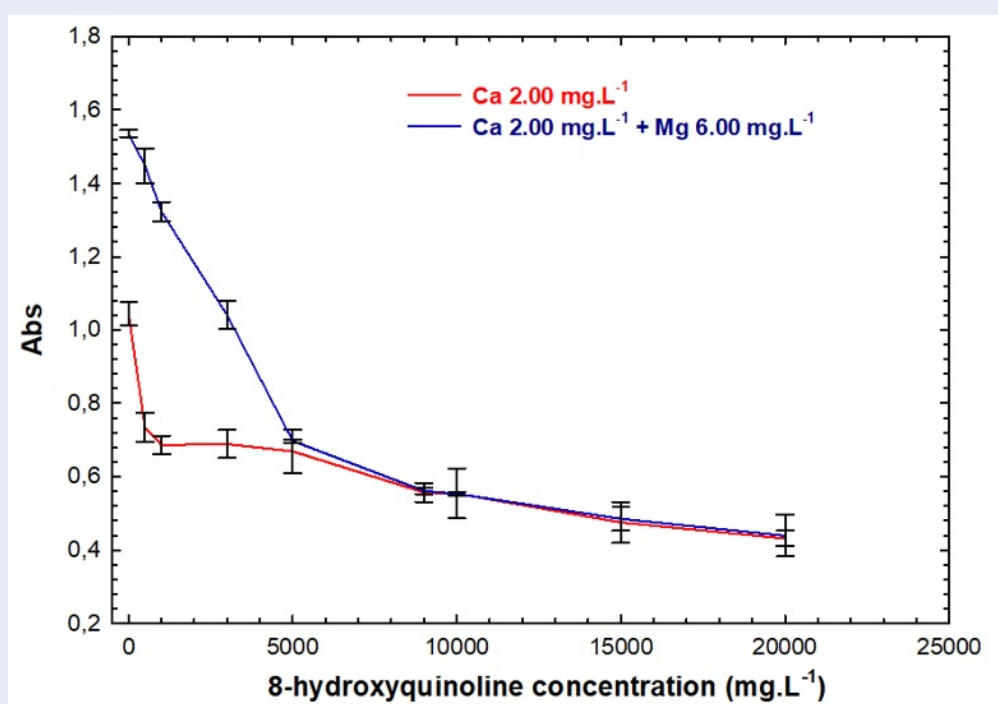


Figure 5: Effect of the magnesium 8-hydroxyquinoline masking agent concentration. The distinction of absorption of complexes was shown between Ca of 2 mg. L⁻¹ (red curve) and the mixture of Ca 2 mg. L⁻¹ + Mg 6 mg. L⁻¹ (blue curve) solutions. The error bar is standard deviations.

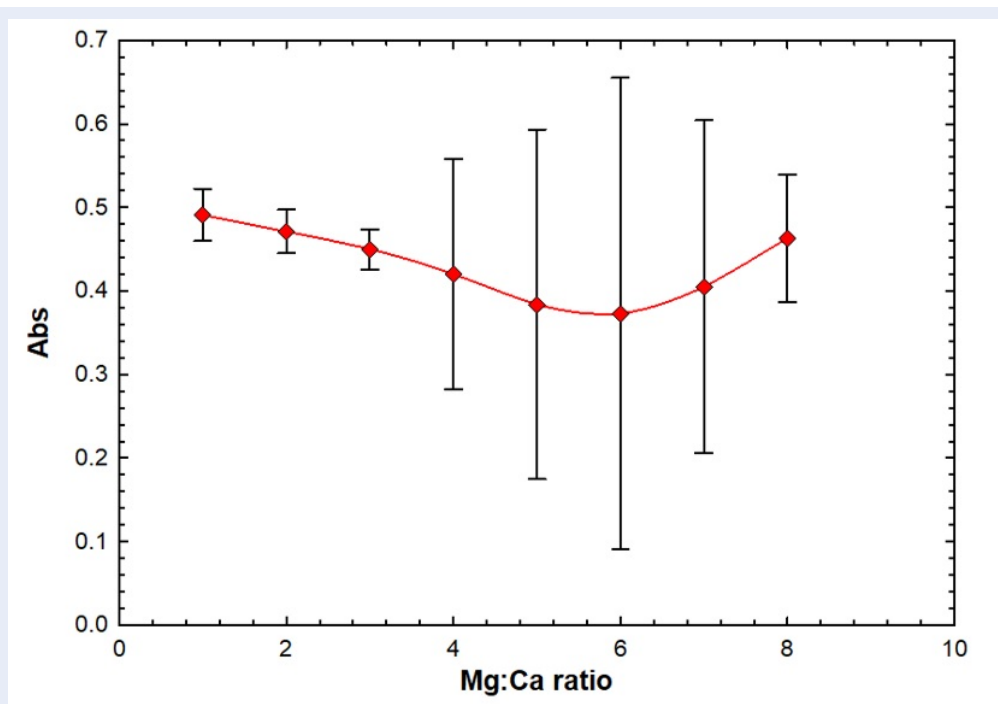


Figure 6: Efficiency of 8-hydroxyquinoline in removing magnesium interference at different Mg:Ca ratios. The change in absorption of complexes within the Mg:Ca ratio range of 1:1-8:1 is displayed (the red curve). The error bar is standard deviations.

Table 3: Ca²⁺ and Mg²⁺ contents in the three seawater samples analyzed using the o-CPC and F-AAS methods.

Method	Samples No.	CCa ± SD	CMg ± SD
		mg.L ⁻¹	
o-CPC	1	344 ± 16	1,339 ± 28
	2	263 ± 5	1,014 ± 23
	3	310 ± 14	1,345 ± 22
F-AAS	1	342 ± 8.7	1,139 ± 29
	2	260 ± 7.0	848 ± 28
	3	347 ± 9.0	1,255 ± 29

*1: Phan Thiet seawater; 2: Quang Ngai seawater; 3: Con Dao seawater

316 for complexation with Ca²⁺ ions was expected
317 (Figure 6).

318 The standard curve model was utilized to quantify
319 Ca²⁺ and Mg²⁺ using the o-CPC method, yielding
320 satisfactory validation results. Comparisons of Ca²⁺
321 and Mg²⁺ contents in seawater samples determined
322 through this method with those obtained via the F-
323 AAS method exhibited favorable results. Statistical
324 analyses, including the F test and t test, revealed no
325 significant differences in variance values ($F_{TN} < F_{0.95}$)
326 or sample mean values ($t_{TN} < t_{0.95}$) between the two
327 methods for determining Ca²⁺ and Mg²⁺ concentra-
328 tions in seawater, ensuring a reliability of P = 0.95.
329 Particularly in the Ca²⁺ analysis, there was notable
330 consistency between the two methods based on the
331 statistical analysis. It can be concluded that the o-CPC
332 method is a reliable alternative for quantifying Ca and
333 Mg in water.

334 Relative to the F-AAS method, the o-CPC method of-
335 fered comparable precision (~2.0–5.0% vs. 2.6–4%)
336 and high accuracy (95–105%) (Tables 1, 2).

337 CONCLUSIONS

338 In this study, the o-CPC method was successfully es-
339 tablished and optimized, offering a rapid means of de-
340 termining Ca and Mg due to two factors. The method
341 is fast, involves an instantaneous colorimetric reac-
342 tion, is simple and efficient, and can be adapted for
343 kit-based testing formats.

344 In this study, the o-CPC method was successfully es-
345 tablished and optimized, offering a rapid means of de-
346 termining Ca and Mg due to two factors: the method
347 exhibits high speed with an instantaneous colorimet-
348 ric reaction, and the procedure is simple, efficient, and
349 can be adapted for kit-based testing formats.

350 Notably, it is a straightforward and dependable tech-
351 nique for assessing Ca²⁺ and Mg²⁺ concentrations in
352 complex matrices such as seawater or saline solutions.

Comparison with the reference method (F-AAS) 353
demonstrated its suitability for analyzing standard 354
samples exhibiting similar characteristics. The o-CPC 355
method is promising for the rapid assessment of Ca²⁺ 356
and Mg²⁺ contents in aqueous solutions and for effi- 357
ciently evaluating the efficacy of water treatment tech- 358
nologies. Such applications represent valuable contri- 359
butions to environmental treatment research. 360

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365 AUTHOR CONTRIBUTION

Tran Duong Nhut and Tran Nhat Quang carried out 366
the experiment with technical support from Hai Son 367
Truong-Lam. Tran Nhat Quang wrote the manuscript 368
in consultation with Hai Son Truong-Lam 369

370 CONFLICT OF INTEREST

The authors declare that there are no conflicts of in- 371
terest related to this study. 372

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