Fourth Derivative Spectrophotometry in Simultaneous Determination of Calcium and Magnesium in Mixture

Moadh .A. AL-Hajaar , M.S. AL-Enizzi and S.Mahfoodh

Dept.of Basic Science, College of Agriculture and Forestry, Mosul University, Mosul, Iraq (Received 10/9/2007, Accepted 30/4/2008)

Abstract

The study included determination of calcium and magnesium in mixture by using 4th–derivative spectrophotometry. The method is based on the red complexes formed by both metal ions with **PURPURATE** (**MUREXIDE**) indicator (violet in colour)in alkaline ethanol – water mixture .The range of application were between 0 to 2.4 μ g / ml and between 0 to 1.4 μ g / ml at 502 &465 nm for calcium and magnesium respectively. A statistical evaluation of the experimental results was reported.

Introduction

The absorbance peaks encountered in UV / Visible spectrophotometry are normally broad due to the overlap of the many molecular transitions taking place in the sample . thus, in some situations; for instance the precise determination of λ max for a broad peak or, precise quantitative and qualitative information can be satisfied. the systematic errors encountered with background or sample turbidity can cause similar problems, unless care is taken prior to sample measurement .Many methods have been proposed to overcome or reduce these problems Unfortunately, they all suffer from numerous analytical Compromises. The more relevant option can be classified into four general groups ⁽¹⁾:

- 1. Standard Addition⁽²⁾
- 2. Measurement by:
- a . Peak Wavelength shift. (1)
- b.Difference spectrophotometry.⁽⁴⁻⁶⁾
- c-Multiple Wavelength easurements⁽¹⁾
- 3. Mathematical correction
 - a. Allen Correction
 - b. Morton and Stubbs
 - c. Glenn
- 4. Instrumental ⁽⁴⁾

5. Derivative spectroscopy.⁽⁵⁾

The spectrum of any compound may be considered as a mathematical function relating the absorbance to the wavelength of a particular compound. thus $A = f(\lambda)$. The function of includes all parameters which affect the absorbance of the compound such as the molar absorptivity of the compound at all wavelengths in the spectrum . mathematically ,it is a function like any other ,and can be differentiated, to other curves such as d A/d $\lambda_{\rm d}^2 A/d \lambda^2$, and so on these curves are known as derivative spectra, those indicated are the first and second derivatives⁽⁵⁾. The first derivative of any function shows the rate of change of that function. In spectroscopic terms, the first derivative shows the rate of change of absorbance with respect to wavelength. the second derivative shows the rate change of the first derivative, again with respect to wavelength. the process may be repeated as often as we like to obtain higher and higher order derivative spectra. Most UV/Visible spectra may be considered to be made up of a series of overlapping Gaussian peaks. It is, therefore, of interest to consider What happens to Gaussian

function when it is differentiated Figure 1shows that



Fig. 1: Idealised representation of $1^{st} / 4^{th}$ derivative on Gaussian peak.

The points to note from this figure are (7)

a. The first derivative crosses the zero axis at same wavelength value as the maximum of the original peak (Y) (zero-order). This is true for any peak shape ,and is often as a way to determine the position of the peak maximum.

b. The second derivative spectrum has a negative peak (Y) corresponding to the

position of original peak maximum.

c. The second derivative spectrum has two 'satellite ' peaks (X1 and X2) one on

each side of the main negative peak.

d.The central, negative peak of the second - order spectrum is narrower than the original peak.

These facts may be generalized for higher order spectra. almost straight line and resolve the minor components The central zero-crossing position remains in live with the more sharply as peak minima with second - order main peak position for all odd order spectra, this peak will derivative and maxima with fourth-order derivative. As alternate in sign from negative to positive as each higher stated previously, increasing derivative orders will order spectrum is calculated . the number of satellites will increase the apparent band sharpening and will, therefore increase with each even-order spectrum, provide an increase in analytical sensitivity up to a

there being four in the fourth derivative at the same time the peak will become narrower and narrower.

Because odd-order spectra do not have any features corresponding to the original peak, these tend not to be used as much as even-order spectra. first derivative spectra are sometimes used because they are simple to obtain and tend not to suffer from excess noise. Second and fourth-order spectra are used most often . Higher orders tend to become very complex ,and as the derivative order increases noise becomes more of a problem .

The representation of the third derivative shows an inverted sense of the first derivative the peak of the zeroorder curves still being at the zero-crossing. however, the use of the fourth derivative produced peak at maximum absorbance .It also exhibits a further sharpening of the peak band. For this application ,the use of second and fourth derivative gives considerable advantage. When small peaks or shoulders appear on the side of a sloping major component back ground, increasing derivative orders will progressively flatten the major peaks to almost straight line and resolve the minor components provide an increase in analytical sensitivity up to a fourth-order derivative and to lesser extent, beyond In recent years, the use of derivative spectrophotometry (DS) has been become more practical owing to the increase in the resolving power of the analytical instrumentation and easier access to microcomputers with appropriate software, which allows the almost instantaneous generation of the derivative spectra⁽⁶⁻⁷⁾

.This facility in the collection and treatment of the spectra has allowed analysis of multi-component mixture of analyte with overlapping spectra . Young, Sweet, and Baker⁽⁸⁾have studied the simultaneous

Young, Sweet, and Baker⁽⁸⁾have studied the simultaneous spectrophotometric determination of calcium and magnesium by *Eriochrome Black* – *T*. depends on obtaining absorbance measurement at a wave length of 630 nm. and at pH of both 9.5 and 11.7 .On the basis of 43 known aqueous mixture of calcium and magnesium .

Tammeline and Mogensen ⁽⁹⁾ have investigated the 1 to 1 complex between calcium and **murexide** in basic medium ranging in colour from yellow-orange to red.

Al-Sindy have studied the simultaneous spectrophotometric determination of calcium and magnesium by first and second derivative spectrophotometry using Bromopyrogallol Red (BPR).⁽¹²⁾

Experimental

Apparatus :

A shimadzu UV/Visible recording spectrophotometer (Model UV-160) and 1-cm quartz cells were used for the normal and derivative spectrophotometric measurements. **Reagents :**

All experiments were performed with analytica reagent grade chemicals and doubly distilled water was used through out .

Solution: 0.1 N NaOH (only) to get pH 12

Calcium Solution, 0.001 M (40 ppm). A 0.0100 gram sample of calcium carbonate solution (Fluka Co. 99.9 %) was dissolved in 5ml of concentrated hydrochloric acid and the solution was diluted to 100 ml.

magnesium Solution, 0.001 M (24ppm). A 0.0040 gram sample of magnesium oxide solution (Merk Co. 99.7%) was dissolved in 5 of concentrated hydrochloric acid and the solution was diluted to 100 ml.

Dye Solution,0.001 M. A 0.0284 –gram sample of <u>pure</u> <u>& dry</u> purpurate ion(murexide)is dissolved in 75 ml. of pure ethanol ,and diluted with 25 ml. of water, store in refrigerator .This solution should be use able for ten days at 5 $C^{o(10)}$

Interfering ion solution ,1000 ppm . A 1000 μ g / ml solution of each ion prepared in double distilled water . Results and Discussion

Spectrophotometric Measurements

Fig. 2 shows the absorbance spectrum of magnesium only which has an absorption maximum at 465nm. The corresponding spectrum of the calcium at 502 nm.

Study of optimum condition *Effect of Reagent Amount*

The influence of the concentration of murexide reagent on maximum formation of the colored complex on each of0.001M magnesium and 0.001 M calcium at pH11.3 was investigated and the results illustrated.

Table 1.Effect of reagent concentration on absorbance									
Ml of 0.001 M murexide	1	2	3	4	5	λ max (nm.)			
Absorbance of magnesium	0.231	0.381	0.498	0.584	0.564	465			
Absorbance of calcium	0.269	0.604	0.766	0.746	0.731	502			
Absorbance of Ca ^{II} &Mg ^{II}	0.251	0.347	0.617	0.705	0.660	482			

 Table 1:Effect of reagent concentration on absorbance

A 4 ml of the reagent was selected because of the highest absorbance .

Effect of Buffer concentration :

sodium hydroxide as buffer was chosen to adjust the pH of the final reaction mixture, The results were shown in Table 2; using 4 ml of reagent

Ml of 0.1 M NaOH	0	0.25	0.5	0.75	1	2	3	4
Absorbance of magnesium	0.000	0.201	0.299	0.252	0.183	0.133	0.035	0.008
Absorbance of calcium	0.040	0.321	0.680	0.692	0.704	0.672	0.718	0.678
Absorbance of Ca ^{II} &Mg ^{II}	0.341	0.415	0.817	0.792	0.768	0.759	0.731	0.700
λ max (nm.) for mixture	472	475	484	485	488	493	489	482

 Table 2 :Effect of buffer amount on absorbance

A 0.5 ml of sodium hydroxide was selected for the subsequent experiments due to magnesium is precipitated in strong alkaline solution .

Order of addition : Both metal ions (M),mixed with reagent (R) and buffer solution (B) which are shown in Table 3 by using zero order.

Table 3	3:Order	of addition	of the reagents
---------	---------	-------------	-----------------

Reaction component	Order number	Absorbance
M+R+B	Ι	0.816
M+B+R	II	0.302

Order I was selected for the subsequent experiments

Quantitative and Qualitative Resolution of Multi-Component Systems:

One of the classic analytical problems for any researcher in the field of UV/Visible spectrophotometry is the resolution of a number of components in a mixture, particularly when the absorbance of those component is extremely variable and/or components with naturally narrow absorption bands. The problems is to resolve small shoulders or peaks from the total spectrum.**Fig.2** (A and B)shows the idealized representation of 4th derivative in the mixture



Fig.2 a,b: shows fourth- derivative absorption spectra of the complexes of Mg^{II} and Ca^{II} with murexide and a mixture of both complexes .It can be seen two closeness overlapping spectra (+ ve maxima) of two ions at 465 and 502 nm.. respectively.

Recommended procedure and calibration curve :

After establishment of the optimum reaction conditions , a calibration curve was constructed by transferring increasing volumes of 24 μ g / ml and 40 μ g / ml of magnesium, calcium respectively into 25-ml volumetric flasks to cover the range 0 to 2.4 μ g / ml for magnesium and 0.16 to 3.2 μ g / ml for calcium, then 4 ml of 0.001 M ammonium purpurate (Murexide) reagent and 0.5 ml of 0.1 M sodium hydroxide. Dilute to volume by double distilled water and read the absorbance immediately against a blank, A plot of absorbance versus determinant concentration shows that Beer's law was obeyed over the concentration 0 to 1.4 and 0.16 to 2.4 for Mg and Ca respectively.



Fig. 3:Calibration curve of magnesium and calcium from 0 to 1.4 and 2.4 ppm respectively

Effect of each ion to other :

To an aliquot of solution containing 1 ml of 0.001M calcium(1.6 ppm), various amount of 0.001M magnesium were added, then 4 ml of 0.001 M reagent, the mixture was diluted in to 25 ml with double distilled water . The

absorbance was measured at wavelength range487 nm. (*for total spectrum of mixture magnesium and calcium* at zero order derivative), and at 470 nm,502 nm (*for each magnesium, calcium respectively by using fourth order* derivative). The results are summarised in Table4 a .

Concentration Mg ^{II} (ppm)	Absorbance of Mg ^{II} at zero order	Absorbance of mixture at zero order	Absorbance o mixture using f deriva Mg ^Π λ max =465	f each ion in fourth order ntive Ca^{II} $\lambda max = 503$	Absorbance of Mg ^{II} only using 4 th order	Recovery of Mg ^{II}
					λ max =465	
0.10	0.157	0.638	0.000	0.122	0.070	0.17
0.24	0.222	0.672	0.000	0.122	0.095	0.30
0.48	0.377	0.736	0.038	0.105	0.137	0.61
0.72	0.477	0.838	0.073	0.093	0.173	0.80
0.96	0.592	0.915	0.108	0.081	0.215	0.95
1.44	0.823	1.027	0.170	0.061	0.270	1.33
1.92	0.970	1.171	0.245	0.031	0.345	1.60
2.40	1.057	1.232	0.315	0.021	0.383	2.15
Slope	2.40		7.08		7.16	
\mathbf{r}^{2}	0.97		0.99		0.99	
Intercept	-0.37		0.19		-0.48	

Table 4 a :Effect of calcium on calibration curve of magnesium at fourth order derivative

From the above table shown that calcium is affected on magnesium at zero order ,but it eliminated in fourth derivative. The same study was applied for the effect of magnesium in presence of calcium, which summarized in Table 4b .

	Table	4 b	: Effect	of ma	agnesium	on calib	ration cu	irve of	calcium	at fourt	h order	deriva	ative
--	-------	-----	----------	-------	----------	----------	-----------	---------	---------	----------	---------	--------	-------

Concentration of Ca ^{II}	Absorbance of Ca ^{II}	Absorbance of	Absorbance of mixture using deriv	of each ion in fourth order ative	Absorbance of Ca ^{II} only using 4 th	Recovery of Ca ^{II}
(ppm)	at zero rder	mixture at			order	
		zero order	Mg		at	
			λ max = 465	λ max = 502	$\lambda \max = 502$	
0.16	0.088	0.656	0.215	0.000	0.028	0.43
0.40	0.186	0.595	0.177	0.015	0.045	0.68
0.80	0.369	0.667	0.155	0.035	0.074	0.96
1.20	0.549	0.814	0.108	0.060	0.100	1.17
1.60	0.638	0.904	0.085	0.080	0.123	1.43
2.40	0.831	1.130	0.025	0.130	0.170	1.96
Slope	2.89			17.3	15.7	
r ²	0.97			0.99	0.99	
Intercept	-0.188			0.166	-0.324	

In order to eliminate the effect of interference of each ion to other ,it will be introducing a term called **inter element correction factor (IECF)**. The value of effect of calcium on magnesium equal (1.3) and for magnesium on calcium equal (0.35), then applied this equation ⁽¹¹⁾:

The true value of magnesium(from table 4a) = (1.3* absorbance of calcium effected at any point + absorbance of magnesium with interference)* Concentration of magnesium from calibration curve/absorbance of magnesium with out interference

For example :

(1):Determination of pure magnesium in mixture :
From table 4a by using 4 th derivative spectrophotometer
(1.3 * A _{Ca} + A _{Mg} with interference)*
C Mg from calibration curve
True value of Mg II in ppm=

A Mg with out interference

Absorbance of Calcium interfere = 0.081Absorbance of Mg in mixture = 0.108Concentration of Mg at this point = 0.96 ppm Absorbance of Magnesium free = 0.215

<u>Solution:</u>

(1.3 * 0.081 + 0.108) * 0.96

Mg II (ppm) = -----= 0.9524 ppm 0.215

Recovery of Mg II= 0.95 / 0.96 *100 = 99.2 % (2):Determination of pure calcium in mixture : From table 4b by using 4th derivative spectrophotometer

 $(\ 0.35 * A_{Mg} + A_{Ca \ with \ interference})_{*} \\ C_{Ca \ from \ calibration \ curve} \\ True \ value \ of \ Ca \ II \ in \ ppm= -----$

A Ca with out interference Absorbance of Mgnesium interfere = 0.108

Absorbance of Ca in mixture	= 0.060
Concentration of Ca at this point	= 1.2 ppm
Absorbance of Calcium free	= 0.100
Solution:	
(0.35* 0.108 + 0.06)*	* 1.2
Ca II (ppm) =	= 1.173 ppm
0.100	
Recovery of Ca II = 1.17 / 1.2 *100	= 97.8 %
Application	
Determination of magnesium and c	alcium in drinking
<u>water :</u>	

The proposed method for determination of two ions without separation in drinking water ,the successful results were obtained in Table 5.

Determination of magnesium and calcium in dolomite A0.1 g of dolomite was wet digestion in acidic medium ,the sample was cooled and filtered up to 25 ml distilled water,then 0.25 ml of aliquot was taken for determination of Mg and Ca in a mixture by derivative spectrophotometer, Table **5** show the results .

<u>Determination of magnesium and calcium in</u> <u>paracetamol tablet</u>

The above procedure is applied.

Sample	Atomic Absorption Spectrophoto- -metry		4 th. Derivative Spectrophoto- ohoto- ry		Recovery %		Complex formation titration with EDTA		Recovery %	
	Mg ⁺²	Ca ^{+ 2}	Mg ⁺²	Ca ^{+ 2}	Mg ⁺²	Ca ^{+ 2}	Mg ⁺²	Ca^+_2	Mg ⁺²	Ca ^{+ 2}
Drinking Water (ppm)	35	40	40	36	Accepte d	Accepte d	24	24	Accepte d	Accepte d
Dolomite %	12.4	21.6	13.1	20	105	92.5	13.8	22	111	101
Paracetamol %	≤0.5	≤0.5	0.4	0.5	Accepte d	Accepte d	0.5	0.5	Accepte d	Accepte d

Table 5: Determination of magnesium and calcium in various samples.

References

1. C.T. Cottrell and V.M. Masters, Derivative and Log Spectrophotometry Ph.D. Pye Unicam Ltd ,Cambridge (1989).

2.C.Mahr,Z.Anal.Chem.,241,133,(1968).

3. N.W.Teitz ,Fundamentals of Clinical Chemistry, Saunders Co., Philadelphia (1976).

4. S. Shibata ,M. Forokawa and K.Goto, Anal . Chim .Acta.,46,271,(1969).

5.A.E.Martiin,Spectro.Chim.Acta,14 97,(1959).

6 A. Y. El Sayed and N.AL-Salem, J . Anal .Sci.,21, 605, (2005).

7. I.. A. AL-Obayidee, Derivative Spectrophotometry for Drugs, Msc Mosul University, College of Science., Chem. Dept .,(2007).

8. A .Young, T. R. Sweet , and B.B. Baker , Anal. Chem 27,3,356, (1955).

9.L.E.Tammelin and S. Mogensen, Acta Chem. Scand., 6,988 (1952).

10.F.W.Diggins, Analyst , 80, 401 (1955) .

11.Operation Manual for Inductively coupled plasma instrumental , Thermo Jarrell Ash Corporation ,Section 3 (1987).

12. L.J. Al-Sindy, Determination of calcium and magnesium using Molecular Absorption Flow – Injection and Derivative Spectrophotometry in Aqueous Solution, PhD., Dohuk University, College of Medicine, (2002).

تقدير الكالسيوم والمغنيسيوم في المزيج بإستخدام مطياف المشتقة الرابعة

معاذ عبد الله الحجار و محمد سالم العنزي و سهير محفوظ

شعبة العلوم الأساسية ، كلية الزراعة والغابات ، جامعة الموصل ، الموصل ، العراق (تاريخ الاستلام: 10 / 9 /2007 ، تاريخ القبول: ٣٠ / ٤ / ٢٠٠٨)

الملخص

شملت الدراسة تقدير الكالسيوم والمغنيسيوم كخليط باستخدام مطيافية المشتقه الرابعه واعتمدت الطريقه على تفاضل الامتصاص مع الطول الموجي الى حد الدرجه الرابعه والحصول على ذروتين منفصلتين حادتين باستخدام كاشف الميروكسايد والحصول على معقد احمر اللون في وسط قاعدي من الكحول والماء واللذان يعطيان اعلى شدة امتصاص عند الطول الموجى٥٠٢ و ٤٦٥ نانوميتر على التوالى وضمن مدايات٠-٢,٤ و٠-١,٤ جزء بالمليون.