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Spectrophotometric Determination of Trace Co (II) with Naphthylglyoxalaldoxime as a newly Synthesized Reagent.

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ABSTRACT

A simple, sensitive and accurate spectrophotometric method for the determination of Co(II) using newly synthesized reagent, 2-(naphthalen-2-yl)-2-oxoacetaldehyde oxime, has been developed. The reagent, 2-(naphthalen-2-yl)-2-oxoacetaldehyde oxime (2NGA), was synthesized based on the reaction of 2-acetyl naphthalene with amyl nitrite in sodium methoxide and reflux at 40°C for 50h. 2NGA reacts with Co²⁺ to form stable yellow complex in basic media which is easily extractable with chloroform. Beer's law was obeyed over the concentration range of 1-15 µg/ml with $r^2 = 0.9990$ and RSD of slope 0.822. Within-day and between-day precision and accuracy values were less than 2.6%. Finally, the method has been applied to a vit B12 product successfully and the results compared with atomic absorption method. The results showed that there was no significant difference between two methods.

Keywords: Co(II), spectrophotometric determination, 2-(naphthalen-2-yl)-2-oxoacetaldehyde oxime.

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INTRODUCTION

Cobalt is known to be essential at trace levels to man, animals and plants for metabolic processes. Some cobalt compounds, such as vit B12 (cyanocobalamin), are noted for their biological activities. Presence of 0.13 to 0.30 mg/kg of cobalt in soils markedly improves the health of grazing animals (1,2).

Cobalt is indicated to be either essential or toxic depending on its concentration range. Lack of the element can induce some disease while it is harmful and deleterious for overtaken. Toxicological effects of large amounts of cobalt include vasodilation, flashing and cardiomyopathy (3,4).

A literary survey reveals that there are several techniques and methods for the determination of cobalt in different samples including atomic fluorescence (5), atomic absorption (6,7), chromatography (8), x-ray fluorescence and spectrophotometry(9,10). Most of these techniques have various limitations of high cost, tedious preliminary separation techniques and excessive use of organic solvents. Among these techniques, visible absorption spectrophotometry using chromogenic reagents represents the most convenient technique because of the availability of instrumentation, simplicity, speed, precision, accuracy and low cost.

Many chromogenic reagents have been reported for trace element determination in different samples (11-15). 2NGA is a new chromogenic reagent recently synthesized in our lab for cobalt determination. The goal of the present study is focused on the synthesis of the new chromogenic reagent, 2NGA, and developed an accurate method for cobalt determination.

EXPERIMENTAL

Materials:

All chemicals were of analytical or HPLC grade and were used without further purification. Cobalt nitrate, tartaric acid and sodium hydroxide were prepared from Fluka. Chloroform, methanol hexane and dichloromethane were purchased from Merck. The reagent, 2-naphthylglyoxalaldoxime, was synthesized in our lab.

Equipments:

The spectrophotometric measurements were carried out using Shimadzu 160 A UV/Vis spectrophotometer. Melting points were determined on a Kofler hot stage apparatus and are uncorrected. The IR spectra were obtained using a Perkin-Elmer Model 781 spectrograph. The ¹H-NMR spectra were obtained on a Varian 400 Unity Plus and chemical shifts (δ) were determined in ppm relative to internal tetramethylsilane. Mass spectra were obtained on a Finnigan MAT TSQ 70 spectrometer at 70 eV.

Solutions:

A solution of 100 μ g/mL was prepared by dissolving appropriate amount of cobalt nitrate in distilled water. The working standard solutions were prepared by appropriate dilution of the stock standard solution. A buffer solution (pH = 9.1 mol/L) was prepared by mixing of boric acid and sodium hydroxide. 2NGA solutions (0.002M) were freshly prepared by dissolving the accurate weight amount in sodium hydroxide (1 M).

Synthesis of 2-naphthylglyoxalaldoxime

2.3 g sodium metal was added to the 50 mL anhydrous ethanol (50 mL) in an ice bath. After stirring for 2 hours 2-acethylnaphthalene (MW=170 g/mol)(9.8 g) was added to the mixture followed by drop wise addition of amyl nitrite under anhydrous condition.

The mixture was stirred at 40 °C for 50 h and after addition of water the mixture was filtered. The filtrate was made acidic by addition of concentrated HCl and then the reagent was extracted with ether. The organic layer was dried over MgSO₄ and the solvent was evaporated to give the yellow precipitate. Crystallization of the precipitate from ethanol/water 50/50 afforded 6.2 g (54%) of the reagent 2-naphthylglyoxalaldoxime (MW=199).

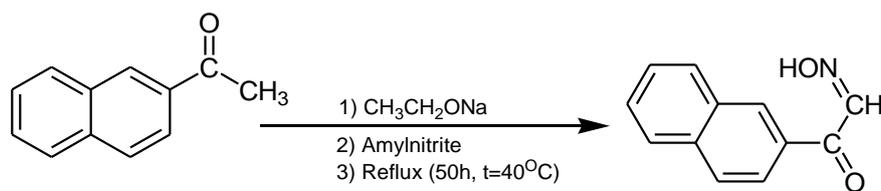


Fig 1. Synthesis of 2-naphthylglyoxalaldoxim

Mp = 89-92°C

yield = %54

IR(KBr) ν Cm^{-1} 3335.9, 3227, 3184 (NH), 3033, 2850(OH), 1619(CO), $^1\text{HNMR}$: 8.67(s, 1H, CH), 8.18(s, 1H, CH), 8.10(d, j= 10 Hz, 1H), 7.98 (d, j = 7.09 Hz, 1H), 7.91 (dd, J = 8Hz, J = 7Hz 2H), 7.64 (t, j = 7.3Hz, 1H), 7.57 (t, j = 7.5 Hz, 1H)

(m/z%) 199 (M^+ , 42), 172 (27), 155(88, 177 (100). 77 (8)

GENERAL PROCEDURE

1 mL of standard solutions containing 1-20 $\mu\text{g Co}^{2+}$, 1 mL of tartaric acid, 3 mL borate buffer (pH = 9) and 2 mL of reagent, 2NGA, 0.002 M followed by 3 mL water was added in a 100 mL separatory funnel to yield the final volume of 10 mL. The resulting complex was extracted with 4, 3 and 3 mL of chloroform. The extracts were collected in a 10 mL volumetric flask and adjusted to the volume with chloroform. The absorbance values of the extracts were measured at 493 nm against a reagent blank.

Preparation of real samples

Contents of 5 Vit B₁₂ ampoules with 5 mL of HNO₃ were heated on a hot plate in a glass beaker to dryness. After that, the sample was cooled, transferred in to a 100 mL volumetric flask and diluted to the mark with water. The resulting solution contains 0.218mg Co^{2+} /mL. working solutions was afterwards prepared through appropriate dilution. Then, the developed method was applied to the final solutions.

RESULTS AND DISCUSSION

The chromogenic reagent 2-naphthylglyoxalaldoxim (2NGA) (Fig. 1) has been synthesized in our lab as a reagent for the determination of cobalt (II) in real samples. It was found that 2NGA reacts with cobalt (II) to form a stable yellow color complex in basic media which is easily extractable with chloroform while 2NGA alone could not be extracted in basic media. So extraction with chloroform was found to be suitable for cobalt determination using 2NGA as chromogenic reagent. Absorption spectra of Co(II)-2NGA complex and the free reagent after extraction with chloroform were made in the wavelength range of 200-800nm are shown in Fig. 2. The Co-2NGA complex showed a maximum absorbance at 410 nm, where the free reagent had no serious interference.

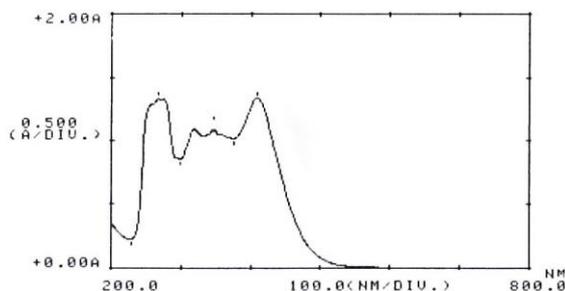


Figure 2. Absorption spectra of Co(II)-2NGA complex in chloroform

Optimization condition

There are some factors that have been obvious effect on the peak shape, sensitivity, concerning pH value and nature of the complex.

In order to find the optimum pH, the influence of pH in the range of 2-13 and sodium hydroxide addition, on the absorbance of Co-2NGA complex was investigated. As seen from Fig. 3, with increasing pH value from 5 to 9 the absorbance of the complex increased. In other words, the absorbance of Co-2NGA complex solutions was decreased above pH 9. The effect of pH surveyed on the free reagent extraction. As it is shown on fig. 4 the absorbance decreased upon increasing pH. Therefore, pH 9 was selected as an optimum pH for further work.

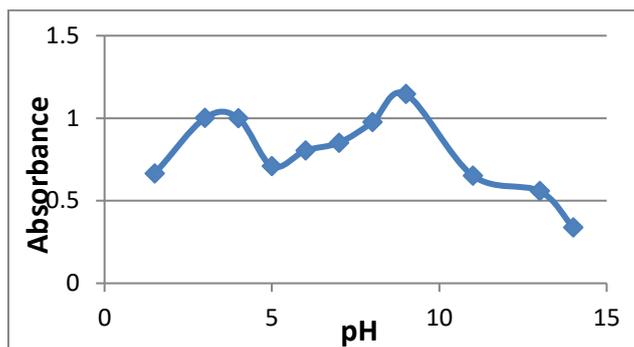


Figure 3. Effect of pH on the absorbance of Co(II)-2NGA complex ($\lambda = 410\text{nm}$)

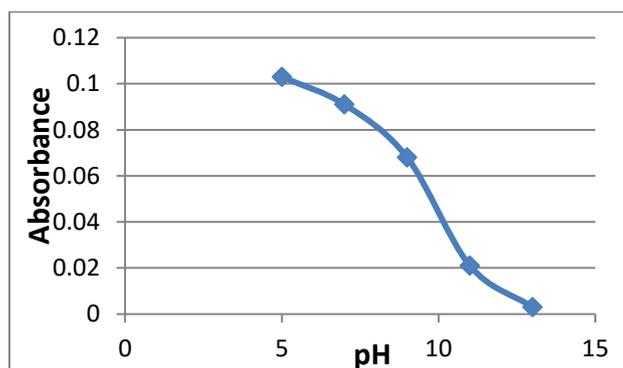


Figure 4. Effect of pH on the 2NGA extraction ($\lambda = 410\text{nm}$)

Stoichiometry of the complex

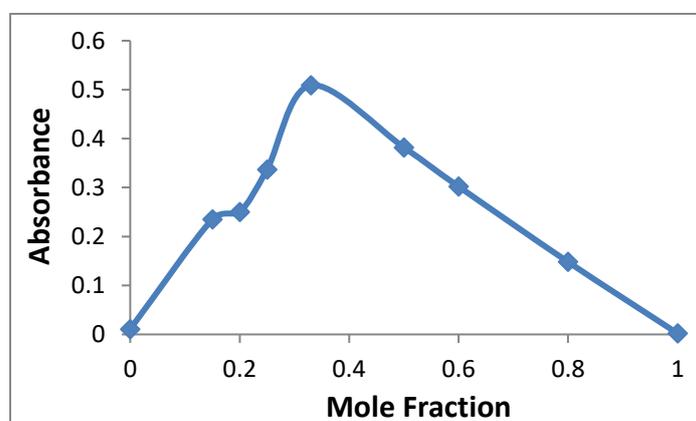


Figure 5. Composition of Co(II)-2NGA complex at 410 nm by continuous variation method

The composition of the complex was determined by continuous variation method using 1.698×10^{-2} M Co^{2+} ($10\mu\text{g/mL Co}^{2+}$). The plot of the absorbance value of Co-2NGA complex solution at 410nm versus the mol fraction of the Co^{2+} , revealed the 1:2 stoichiometry of metal ion: ligand (Figure 5).

The effect of concentration of the ligand, 2NGA, was also studied for the Co^{2+} solution containing 1.698×10^{-2} M. By increasing concentration of 2NGA from 1.698×10^{-2} M to $4 \times 1.698 \times 10^{-2}$ M the absorbance value increased. When the concentration passes beyond the ratio of 4, absorbance was nearly constant (Fig.6). Therefore, in all determinations of cobalt, a small volume of reagent more than this ratio was employed.

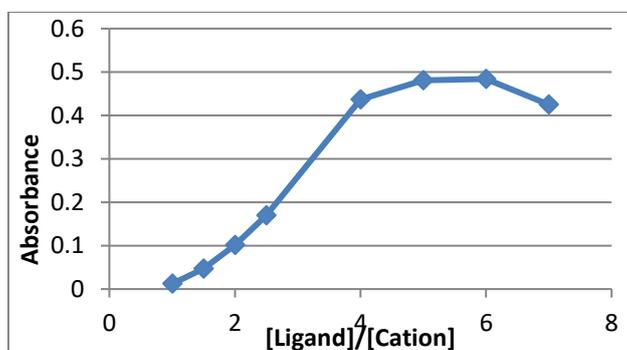


Figure 6. The effect of 2NGA amount on the absorbance at 410 nm

Under the optimum conditions, linear regression equation (intercept and slope) for determination of Co^{2+} was established. The high values of correlation coefficient were 0.9990 indicating the good linearity of calibration curves.

Table 1: Analytical data of calibration curve and molar absorptivity of Cobalt (II) (n=7)

Parameters	Co
Linearity	1-15 ($\mu\text{g/ mL}$)
LOD	0.550($\mu\text{g/ mL}$)
ϵ	$3930 \text{ Lmol}^{-1}\text{cm}^{-1}$
Regression equation	$Y= 0.06516X - 0.02553$
SD of slope	5.4×10^{-4}
RSD of slope	0.822
SD of intercept	0.012
Correlation coefficient	0.9990

All the validation parameters, such as concentration range, correlation coefficient and detection limit (LOD) are summarized in table 1. Accuracy and precision was determined with three replicate determinations at five different concentrations in the calibration range, whose results are provided in table 2. The accuracy is evident from the data as percent error is less than 2.13 in all determinations. Percentage relative standard deviation was less than 2.6%, indicating good precision of the proposed method. The correlation coefficient was calculated to be higher than 0.9990, which is indicative of good linearity.

Table 2. Accuracy and precision data for determination of cobalt in one day (n = 3) and three subsequent days (n = 9).

Added, $\mu\text{g/ ml}$	Within-day (n=3)			Between-day (n=9)		
	Found	CV%	Error%	Found	CV%	Error%
1	1.02 ± 0.02	1.64	1.58	1.03 ± 0.02	2.12	2.57
5	4.96 ± 0.13	2.58	-0.84	4.92 ± 0.10	2.04	-1.55
10	9.80 ± 0.21	2.19	-1.99	9.92 ± 0.18	1.85	-0.85
12	12.11 ± 0.23	1.94	0.91	12.05 ± 0.13	1.07	0.39
15	15.04 ± 0.02	0.14	0.29	15.09 ± 0.10	0.65	0.57

Application to real samples

The proposed method can be used for determination of Co^{2+} in vit_{B12} ampoule, containing 100 mg cyanocobalamine equal to 4.36 mg Co^{2+} . The results achieved by the proposed method were compared with atomic absorption method. As it is shown in table 3, the percentage of recovery is 99.1% and 99.8% for spectrophotometry and atomic absorption method, respectively.

Table 3. Application of the proposed method to the determination of Co(II) in Vit B12 (n = 3).

Co^{2+} in vit _{B12} (mg)	Founded (mg) (spectrophotometry)	%Recovery	Founded (mg) (atomic absorption)	%Recovery
4.36	4.321 ± 0.006	99.1	4.351 ± 0.001	99.8

Relative recovery

To check the effect of excipients on the quantitative analysis, the standard addition technique using four replicates was applied.

The recovery of 98% and relative standard deviation of 0.02 indicates that no interference of other compounds was observed during analysis.

Added Co^{2+} (μg)	Recovery (Mean±SD)
15	%98±0.02

CONCLUSION

The proposed spectrophotometric method using the newly synthesized reagent (2NGA) is demonstrated to be a simple, rapid, convenient and sensitive ($\epsilon = 3930 \text{ Lmol}^{-1}\text{cm}^{-1}$) method for the determination of Co (II). The method shows a low detection limit (LOD) for cobalt.

The results achieved were comparing with those obtained by atomic absorption method and successful results were recorded.

The developed method may be recommended for routine analysis of Co(II) in pharmaceutical and industrial samples.

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