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A GREEN AND ECONOMICAL METHOD DESIGNED FOR DETECTION OF COBALT IONS UTILIZING BENZOHYDRAZIDE AS REAGENT

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Article Info

Abstract

A green, simple, and cost-effective spectrophotometric method for the accurate determination of trace cobal (Co (II)) ions was developed utilizing Benzo hydrazide as a chromogenic reagent in a micellar Triton X−100 medium. The method relied on rapidly forming a light pink Co (II)-BH2 complex at an optimum pH of 9.0. The complex exhibited maximum absorption (λ_max) at 436.2 nm and demonstrated excellent temporal stability, with constant absorbance for up to 140 minutes. The proposed method confirmed a 1:2 metal-to-ligand (M: L) ratio. It conformed to linear maximum concentrations range 0.1 to 5.0µgmL-1 with a high correlation coefficient (R2) of 0.999. The high sensitivity of method is evidenced by a molar absorptivity (ϵ) of 4.13×104Lmol-1cm-1 and a Sandel's sensitivity of 5.3 ngcm-2. The technique was successfully applied and validated by comparing results against certified values and Atomic Absorption Spectrometry (AAS) in various real-world samples, including pharmaceuticals, certified alloy, water, and wastewater.

Keywords:
Spectrophotometric Method, Benzo hydrazide, Reagent, Triton X−100, Surbex-Z

INTRODUCTION

Cobalt ions is a vital micronutrient essential for various biological processes like biosynthesis and metabolism etc. [1]. In living organisms, it is a fundamental component of vitamin B_{12} (cobalamin), which is required for red blood cell formation, certain metalloproteins and neurological function [2]. However, excessive exposure to cobalt ions can lead to severe health risks by generating reactive oxygen species (ROS), including asthma, pneumonia, interstitial lung diseases, cardiomyopathy, thyroid dysfunction, and neurotoxicity [1, 3]. Industrial activities such as metal plating, pigment manufacturing, mining, and battery production often release cobalt into the environment, contaminating soil and water sources [4, 5]. Therefore, the accurate and reliable determination of cobalt at trace levels in environmental, biological, and industrial samples is of significant importance for public health and environmental monitoring [6].

Traditional methods used for cobalt analysis include inductively coupled plasma mass spectrometry, inductively coupled plasma optical emission spectrometry and atomic absorption spectrometry [7-9]. Whenever these techniques provide high sensitivity and accury, these are mostly associated with more operational costs, the requirement for sophisticated instrumentation, complex sample preparation, and non-eco-friendly reagents [10]. Consequently, there is a growing interest in developing simple, cost-effective, and environmentally benign analytical methods for cobalt determination.

Spectrophotometric techniques have gained considerable attention due to their simplicity, low cost, rapid response, and adaptability to routine analysis [11]. The key to an effective spectrophotometric method lies in the development of a suitable chromogenic reagent that selectively forms a stable and intensely colored complex with cobalt ions [12]. Among various ligands, hydrazide derivatives have emerged as efficient chelating agents because of their ability to form coordination complexes through the carbonyl and amino groups [13].

Benzo hydrazide, a simple and environmentally safe organic compound, has been explored as a potential analytical reagent due to its strong affinity toward transition metal ions and its ability to produce colored complexes suitable for spectrophotometric detection [14]. Its non-toxic nature and ready availability make it a promising candidate for green analytical chemistry applications. Furthermore, using benzo hydrazide as a ligand aligns well with principles of green chemistry via lessening the usage of dangerous solvents and decreasing chemical waste [15]. The present study focuses on the development of a green, sensitive, and cost-effective spectrophotometric technique of determining cobalt ions employing benzo hydrazide. Proposed technique not only reduces the environmental footprint of chemical analysis but also offers a practical alternative to expensive instrumental techniques.

Material and Methods

Reagent Preparation

The solution 4×10^{-4} Molar of Benzo hydrazide was prepared adding 0.0544gram in 3.0% Triton X-100 to a 1000 milli liter (mL) measuring flask, containing the minimum amount of ethanol . To make the initial measured Triton X-100 3.0% solution, its 3gram was added to 100 milli liter (mL) of calibrated flagon, and the amount was made with double distilled water [16]. To make the metal stock solution of cobalt, 4.04 grams of $CoCl_2.6H_20$ were added to double distilled water. To make the metal ion stock

solutions, the right amount was dissolved in double-distilled water. For the acidifying procedure, it with a solution of 2% nitric acid. The buffer solutions from pH 1 to pH 10 were prepared following the protocols by addition of appropriate amounts of HCl-KCl equimolar 0.2Molar for pH 1-4, CH3COOH-CH3COONa equimolar 0.2Molar for pH 5-6, KH2PO4-NaOH equimolar 0.1.A 0.025 M sodium borate–0.1 M HCl buffer system was utilized to maintain pH values in the range of 9–10, while molar buffer solutions were prepared to achieve pH values between 6.5 and 8 [17].

Determination of Cobalt in Vitamin-B12 (Neurobion) Injection

Samples of Neurobion injections (Merck, 1.0 mL vitamin B12) were transferred into a round-bottom flask containing 50 mL of solution prepared with 10 mL of a mixed acid reagent composed of sulfuric and nitric acids in a 1:1 ratio. The volume of the samples was almost completely dried out after reacting with heating on a hot plate. The final transparent residue was obtained by agitating the residual sample solutions with nitric acid after they had been The samples were leached with dilute H_2SO_4 and subsequently diluted to the calibration mark in a volumetric flask using a dropwise technique. Afterward, the solutions were reacted with 4×10^{-4} mL of benzo hydrazide at pH 9.0 in 3.0% Triton X-100. Absorbance values of the resulting cobalt–benzo hydrazide metal complexes were then measured. The corresponding analytical data are summarized in Table 6.

Cobalt (II) Ion Detection in Alloy

In separate beakers, alloy samples of JSS 607-6 containing Co(II) (0.1 to 0.5 grams) were treated with 5.0 mL of concentrated nitric acid (HNO_3) for dissolution. Concentrated hydrochloric acid (HCl) and concentrated sulfuric acid (15 mL) were also added. After that, the resultant solution was slowly heated on a hot plate until it had decreased in volume to around 5 mL. Sample solutions were filtered and diluted with 10 milli liter conc. HCl solution, resulting in a ending volumes of 25mL in volumetric flagon. Measurements were made of the co-complex absorbances of the specimens following their reaction with 4×10^{-4} M Benzo hydrazide pH 9.0 in 3.0% Triton X-100. The results are shown in Table 5.

Examination of Cobalt (II) in Tap Water

A tap water sample was collected from the Thari Mirwah region of the Khairpur district. The sample was subsequently filtered through a 0.45 µm membrane filter to remove suspended particulate matter. 1 mL of concentrated HNO_3 was added to acidify the solution and stop precipitation. After adding two milliliters of 4×10^{-4} M Benzo hydrazide, 2 milliliters of pH 9.0 buffer, and 2 milliliters of 3.0% Triton X-100 mixed to the specimen, the Co-complex absorbance was measured. Presents the findings in Table 3 .

Analyzing Co (II) from Environmental Water

One-liter samples of wastewater have been collected from various sites in Khairpur, Pakistan. Filtration and acidification of the specimens were performed by adding 2 mL of 30% concentrated H_2O_2 and 4 mL of concentrated HNO3. After that, 25 mL of solutions were obtained by pre-concentrating the resultant solutions in an oven set to 110°C. Co-complex absorbance was then measured after the solutions of specimens were moved to graduated container and mixed with 2mL 4×10^{-4} M BH, 2mL pH 9.0 buffer, and 2 mL 3.0% Triton X-100. Findings are shown in Table 3

Results and Discussion

Cobalt Spectrophotometric Research with the Triton X-100

Benzo hydrazide [BH] reacted with Co (II) ions at pH 9.0 in an aqueous solution containing Triton X−100 to form a light pink Co-$[BH]_2$ complex.

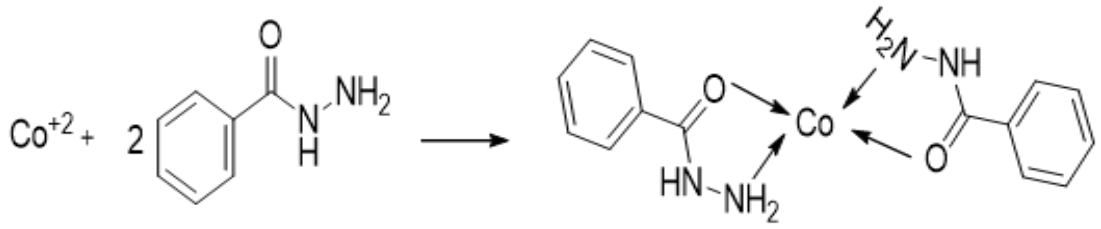

Figure. 1. Proposed Reaction of Benzo Hydrazide Reagent with Co^{2+} Formed [Co (II)-BH_2] Complex

Absorption Spectra of Ligand BH and Co (II)-BH_2 Complex

The initial absorption spectrum of the benzo hydrazide reagent was measured against a solvent blank. Subsequently, the spectrum of the Co (II)-BH_2 complex was recorded against the reagent blank. Analysis of these spectra revealed that the BH reagent showed its maximum absorption at λ_{max} 285 nm, while the Co (II)-BH_2 complex showed its maximum absorption at λ_{max} 436.2 nm as shown in Fig. 2 and Fig. 3.

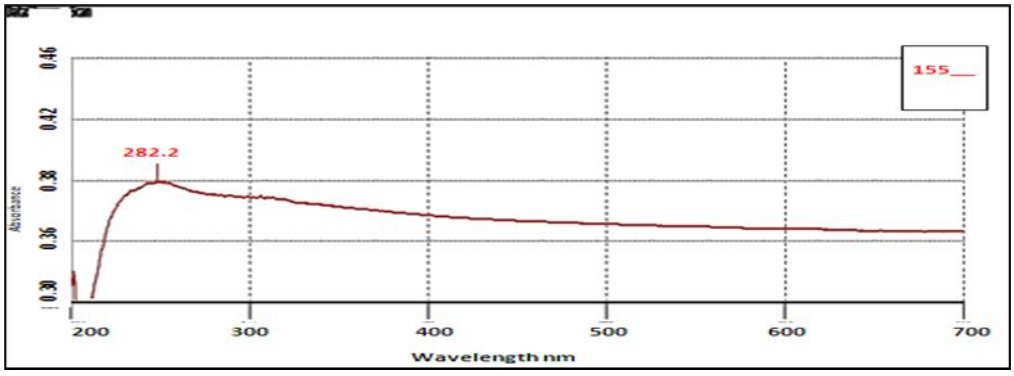

Figure. 2. UV-Vis Electronic Spectra of Benzo Hydrazide at Λ_{max} of 282.2 Nm in Triton X-100

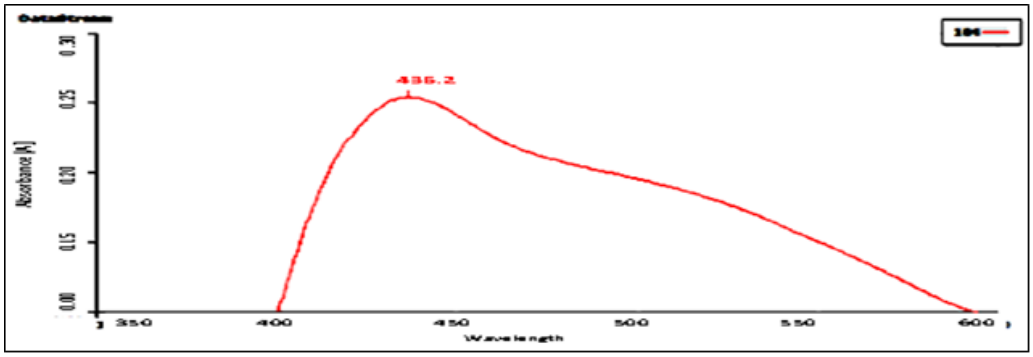

Figure. 3. UV-Vis Electronic Spectra of Co-BH_2 at λ_{max} 436.2 nm in Triton X-100

Effect of pH:

Impact of pH on extraction recovery was assessed while keeping other parameters constant. It was determined that the ideal pH for iron extraction is 9.0, which has been selected for further investigation, as illustrated in Figure 4 and Table 1.

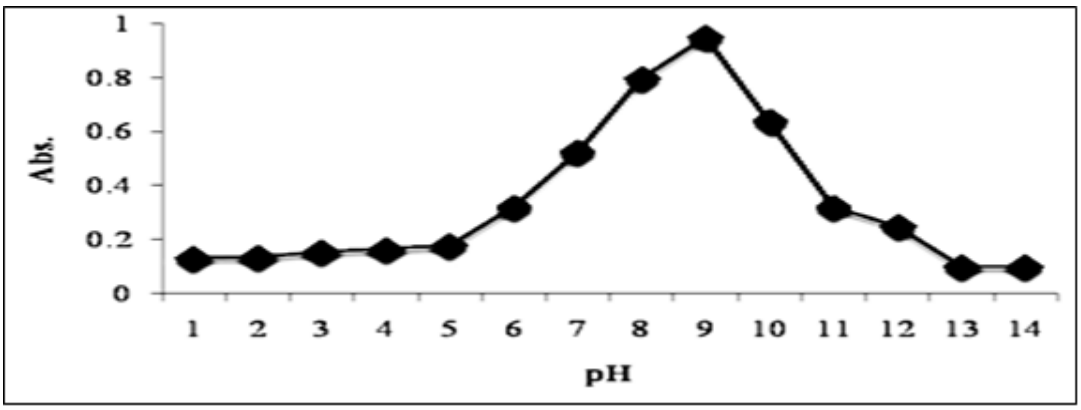

Figure. 4. pH profile of the Co(II)–BH_2 Complex in a 3.0% Triton X-100 Micellar Medium

Ratio of Metal to Complexing Reagent

The metal–ligand complex's stoichiometric composition was ascertained using the molar ratio approach [18]. The analysis revealed a cobalt-to-benzo hydrazide molar ratio of 1:2, indicating the formation of a Cobalt(II)–BH_2 chelate complex, as presented in Table 1**.**

Effect of Concentrations of Triton X-100 and Complexing Reagent BH

Various concentrations of the nonionic surfactant Triton X-100 were examined to evaluate their influence on complex formation. The maximum absorbance was observed when a 3.0% Triton X-100 solution (2 mL) was utilized in conjunction with a fixed metal ion concentration of 2 mg/L.

Effect of Concentrations of Complexing Reagent BH

The complexation of the metal with the chelating agent was affected by the concentrations of the BH ranging from 0.5-8×10^{-4} M. Absorbances were recorded using various concentrations, and the metal chelate maximum absorbance was found at 4.0×10^{-4}M concentration, which was considered the ideal condition and used throughout the study as shown in Fig. 5 and Table 1.

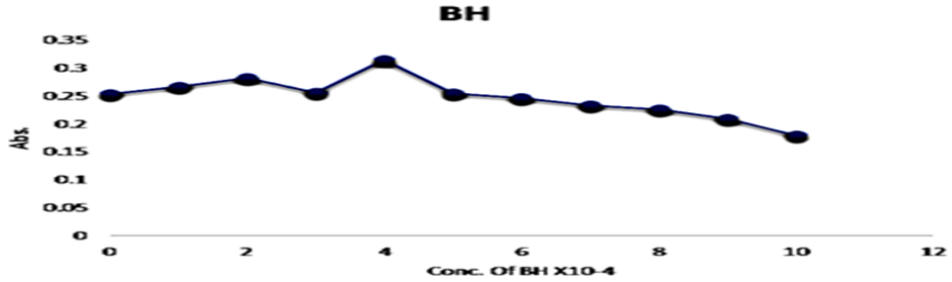

Figure. 5. Effect of Benzo Hydrazide Concentration on the Absorbance of The Complex

Time Influence

The synthesis of metal chelates was investigated; the complexation was rapid, provided a fixed maximal absorption at room temperature, and stayed that way for 140 minutes.

Cobalt(II)-Benzo Hydrazide Calibration

The calibration curve for Co(II) at λ_{max} = 436.2 nm exhibited linear concentrations range 0.1–5.0 mg/mL, with a correlation coefficient (R^2) of 0.9994 and a zero intercept, as illustrated in Figure 6.

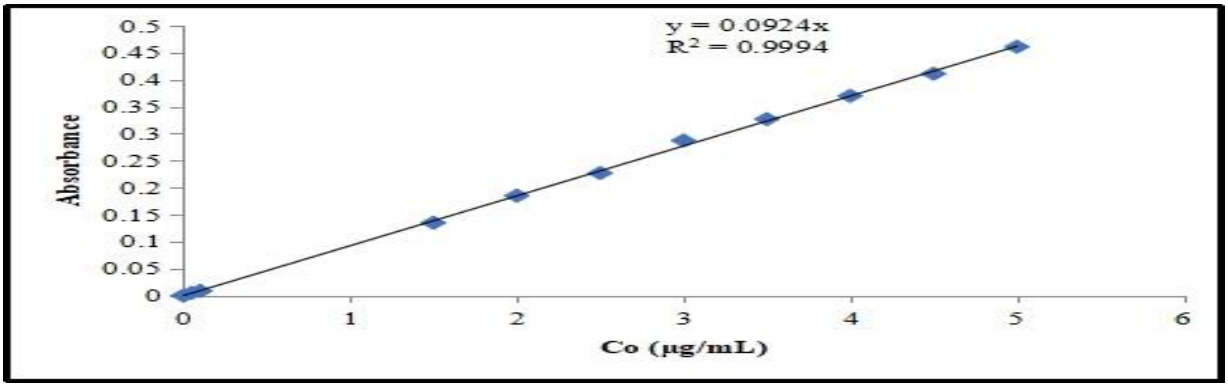

Figure. 6. Calibration Graph of Co(II)-BH_2

Sandell's Sensitivity, Limit of Detection (LOD), and Molar Absorptivity Coefficient

Average molar co-efficient absorption for Co (II) at λ_{max} 436.2 nm was determined to be 4.13×10^4 Lmol-1cm-1 using a linear calibration curve. 5.3 $ngcm^{-2}$ was reported as the limit of detection. According to Table 1, Sandell's sensitivity was found 5.3 ng/cm^2. The outcomes were comparable to those reported in the literature.

Table 1. Optimum Parameters for Co (II)-BH_2 Complex

Parameters	Cobalt
Absorption max: $\lambda_{\_max}$	436.2 nm
Molar absorptivity	4.13×10^4 L mol^{-1} cm^{-1}
pH	9.0
Triton X−100 3.0%	2.0 mL
Benzo hydrazide	4.0×10^{-4} M
M:L Ratio	1:2
Beer's Law range	0.1-5.0 µg/mL
Sandell's sensitivity	5.3 ng/cm
LoD	5.3 ng/mL
R^2	0.9994

Influence of different Ions in Cobalt Detection

Solutions containing various proportions of multiple anions and cations, as well as 1µg of cobalt (II), were prepared using the same method . The interference limit of each ion was investigated by determining the ratio at which a ±2% variation in the absorbance of the complexes was observed [19], as presented in Table 2.

Table 2. Interferences of different anions, cations and salts

Ion / Salt	Cobalt (Tolerance Ratio)
$KClO_3$, Na_2tartarate	1000
Mg(II)	800
NaF	600
Zn(II), Al(III)	100
Cr(III)	30
Cu(II)	10
Cr(IV)	8
Ni(II)	5
Fe(III)	5
Pb(II)	3
Cd(II)	2

Validation of Method

The proposed method was applied for the quantitative determination of cobalt(II) in natural, alloy, pharmaceutical, environmental, and biological samples. The results obtained demonstrated excellent concordance with those measured by atomic absorption spectrometry (AAS), as presented in Table 6.

Existing procedures were contrasted with this one. Compared to earlier methods, the developed suggested procedure has improved Sandell's sensitivity, linear calibration range, limit of detection, and molar absorptivity (Table 7).

Table 3. Percentage Recoveries of Cobalt (II) Mixed to Tap and Waste Water Specimens

Specimens	Cobalt mixed (µg/mL)	Detected with Proposed Method (µg/mL)	Detected with AAS (µg/mL)	% Recoveries
Tap water	2.5	2.3	2.4	92
Wastewater	00	1.00	1.00	
	3.0	3.89	3.90	97.25

Table 4. Co(II) Determination in Pharmaceutical Sample

Samples	Metallic ion	Existing (µg/mL)	Suggested technique (µg/mL)	%Recoveries
Surbex Z (Abbott)	Cobalt(II)	2	**1.94**	97

Table 5. Analysis of Cobalt in Certified Alloys

Certified Alloy	Cobalt present	Cobalt found	% Relative error	% Recoveries
JSS 607-6	14.01 µg	13.98 µg	0.14	99.78

Table 6. Co(II) Determination in Neurobion (Injection) Sample

Samples	Suggested Procedure (μgmL^{-1})	%RSD	AAS Method (μgmL^{-1})	RSD%	%Recovery
Neurobion (Inj.) 21.74 μgmL^{-1}	21.68	0.4	21.69	0.4	99.7

Table 7. Comparison of Investigation Method for Co (II) using Benzo hydrazide reagent

Metal Ion	Methods	References
Co(II)	Triton X-100, ε 3.18×10^4 $Lmol^{-1}cm^{-1}$, Sandell's sensitivity, 2.05 $ngmL^{-1}$, linear range 0.20-3.0 μgmL^{-1}, and D.L 1.68 $ngmL^{-1}$	[20]
Co(II)	CTAB, ϵ 1.89×104 $Lmol^{-1}cm^{-1}$, Sandell's sensitivity $1.89\times10^4 L\ mol^{-1}cm^{-1}$ and 3.1 $ngcm^{-2}$ at λ_{max} 577.8 nm. Linear range 0.12−6.0 μgmL^{-1}	[21]
Co(II)	LoD 0.02 µg/mL, at λ_{max} 425 nm in Triton X-100, ε 2.05×10^4 $Lmol^{-1}cm^{-1}$	[22]

Co(II)	Benzo hydrazide 4.0×10^{-4} M, in Triton X-100 3.0%, Molar absorptivity 4.13×10^{4} $Lmol^{-1}cm^{-1}$, Beer's Law range 0.1-5.0 μgmL^{-1} , λ_{max} 436.2 nm. pH 9.0, Sandell's sensitivity 5.3 $ngcm^{-2}$, DL 5.3 ng/mL, R^2 0.9994, M:L Ratio 1:2	Present study

Conclusion

A green, simple, and cost-effective spectrophotometric method has been successfully developed for the accurate determination of trace amounts of Co(II) using Benzo hydrazide in a micellar Triton X-100 medium instead of old method of solvent extraction. Method is based on the formation of a stable Co(II)-BH_2 complex at λ_{max} 436.2 nm. Complex adheres to linear concentration range 0.1–5.0 μgmL^{-1}. The method demonstrates high sensitivity, as indicated by a molar absorptivity of 4.13×10^{4} $L\cdot mol^{-1}cm^{-1}$ and Sandel's sensitivity of 5.3 $ngcm^{-2}$. Its accuracy and reliability were validated through successful application to real samples—including pharmaceuticals, certified alloys, water, and wastewater—showing strong agreement with certified values and results from Atomic Absorption Spectrometry (AAS). The proposed analytical procedure was successfully employed for the determination of Cobalt ions (II) in natural, alloy, pharmaceutical, environmental, and biological samples.

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