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University Of Thi-Qar
College Of Science
Department of Chemistry



Determination Of Some Heavy Metals In Real Samples Using New Azo-Azomethine Compounds By CPE

A thesis

Submitted to The Council of The College of Science, University of Thi-Qar
in Partial Fulfillment of The Requirement For
The Degree of Doctor of Philosophy in Science/Analytical chemistry

By

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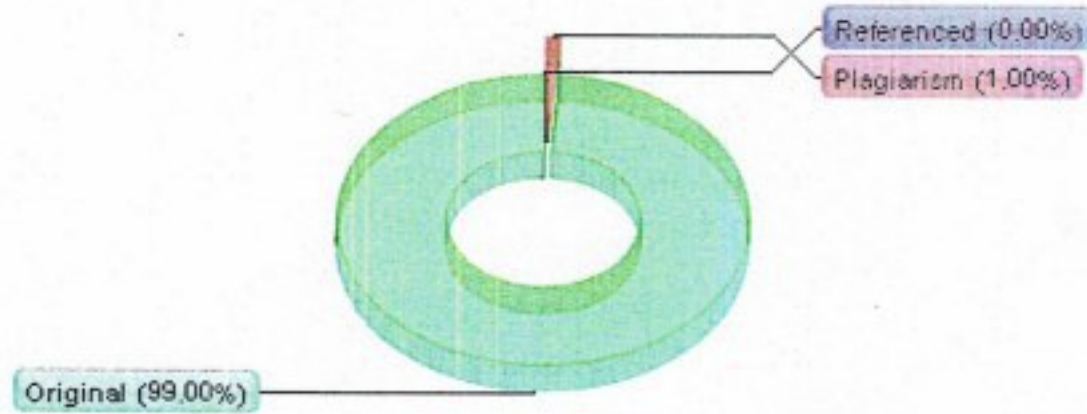
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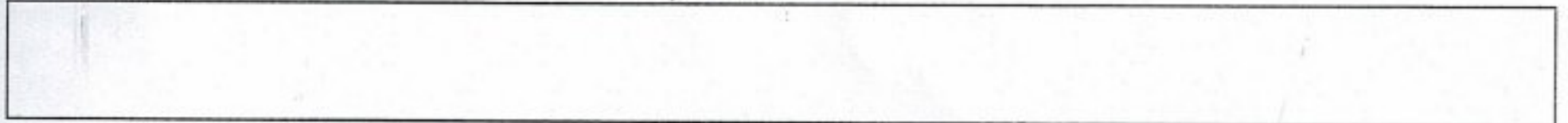
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والصادر بتاريخ 2014/3/31، عند أعداد الرسالة أو الاطروحة الموسومة:

Synthesis and characterization of some Azo-

Azomethine compounds and their using in the determination

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Flame atomic absorption spectrometry after separation and

وبخلافه نتحمل نحن الموقعون أدناه المسؤولية القانونية حال وجود ما
pre concentration with cloud point extraction methodology.
بخالف مضمون الكتاب أعلاه أو ما ينافي أخلاقيات البحث العلمي.

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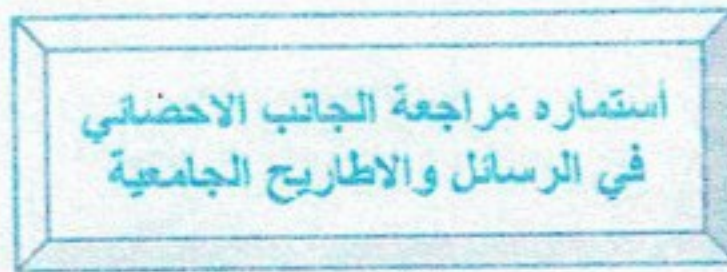
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I certify that this thesis, "**Determination Of Some Heavy Metals In Real Samples Using New Azo-Azomethine Compounds By CPE.**" written by Sajda Sabbar Affat has been prepared under my supervision at the Department of Chemistry, College of Science, University of Thi-Qar (Iraq) in partial fulfillment of the requirements for the degree of doctor of philosophy in Analytical chemistry.

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In view of the available recommendation, I forward this thesis for debate by the examining committee.

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CERTIFICATE

We, the examining committee, after reading this thesis "**Determination Of Some Heavy Metals In Real Samples Using New Azo-Azomethine Compounds By CPE.**" and examining the student **Sajda Sabbar Affat** in its content, find that it is qualified for pursuing the degree of doctor of Philosophy in Analytical chemistry on **(December , 2018) .**

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إهداء



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أبي الغالي
إلى التي هي للعين نور وللقلب فرح وسرور حبيبة الأحباب
إلى التي أعلى الله مقامها فجعل الجنة تحت اقدامها...

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إلى تسابيح الصباح الجميلة المملوءة إشراقةً وبهجة...
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إلى عنوان المحبة الذي رحل عنا عمي الغالي

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List of Abbreviations

SPME	Solid-phase micro extraction
LPME	Liquid phase micro extraction
HLLE	Homogenous liquid liquid Extraction
DLLME	Dispersive liquid-liquid micro extraction
SDME	Single drop micro extraction
CPE	Cloud point extraction
SPE	Solid phase extraction
LLE	Liquid-liquid extraction
SPP	Surfactant-poor phase
CMC	Critical micelle concentration
POE	Polyethoxylated
HLB	Hydrophilic-lipophic balance
PIT	Phase-inversion temperature
TX-100	Triton X-100

HPLC	High performance liquid chromatography
US EPA	Environmental Protection Agency
ICP-MS	Inductively coupled plasma-mass spectrometry
ICP-AES	Inductively coupled plasma-atomic emission spectrometry
GFAAS	Graphite furnace atomic absorption spectrometry
UV-Vis	Ultra violet-Visible
FT-IR	Fourier Transform Infrared
m.p	Melting point
FAAS	Flame Atomic Absorption Spectrophotometric
¹H-NMR	Proton Nuclear Magnetic Resonance
TLC	Thin Layer Chromatograph
hr	Hour
min	Minutes
DMSO	Dimethylsulfoxide
nm	Unit of Nanometer
°C	Degree Celsius
%	Percentage
δ	Chemical shift
ml	Milliliter
ppm	Part per million
KBr	Potassium bromide
CsI	Cesium iodide
HCl	Hydrochloride acid
gm	Gram
M	Molarity
ΔH	Enthalpy change
ΔG	Gibbs free energy change
ΔS	Entropy change
Log	Logarithm
LOD	Limit of detection
LOQ	limit of quantitation
EF	Enrichment factor

PF	Preconcentration factor
CI	Consumptive index
%RSD	Percentage relative standard division
R²	Correlation coefficient
ABS	Absorbance
D	Distribution ratio
OFAT	One-factor-at-a-time
WHO	World health organization
K_{ex}	Equilibrium extraction constant
IDPI	2-(3- indolyl) - 4,5 di phynyl imidazole
CV	Crystal Violet
IPAI	3-(1-(1-H-Indol-3-Yl)-3-phenylallyl)-1H-indole
PPT	Phenanthraquinone monophenyl thiosemicarbazone
ANPAP	Amino-4-(3-nitrophenylazo)pyridine-3-ol
SPAR	Sulfapyridylazo resorcinol
DDTC	Diethyldithiocarbamate
MPMP	2-[(2-mercaptophenylimino)- methyl]phenol
BTANP	2-(benzothiazolyl azo)-4-nitrophenol
MOSDAA	4-methoxy-2-sulfobenzenediazo-aminoazobenzene
1-PTSC	1-Phenylthiosemicarbazide
CPT	Cloud point temperature
PONPE	Poly ethylene glycol mono-4-nonylphenyl ethers
SRP	Surfactant rich phase

Summary

Since its inception in 1985, Cloud-point extraction (CPE) methodology has constituted an important theme in the analytical chemistry as promising procedure for the separation and preconcentration for the metal ions and organic compounds from the complex matrices samples. Nowadays, it has been begins to take a large noteworthy position among the other modern separation methods in scientific research and application on a high level due to its simplicity, rapidity, more precise and cheapness beside an environmentally-friendly method, therefore, this research includes three main chapters .

Chapter One includes introduction of azo-azomethine compounds and their complexes and a view of the different extraction techniques, fundamental principles and potential applications of CPE methodology in analytical chemistry. It also involves a concise review on the latest developments of the method and its applications for the determination of cadmium, copper and cobalt by CPE .

Chapter Two consists of an outline of different instrumental techniques , general apparatus and chemicals used in the present work. The synthesis paths of azo-azomethine compounds (L_1 to L_4) and primary testing of complex formation between metal ions and reagents, determination of $\lambda_{\max}$. As well as optimization of the experimental conditions (volume reagent, reaction time, temperature and pH) and determination of dissociation degree and stability constant, stoichiometry of the complex (Continuous variation method -Job method). In addition

to, the complexes were prepared for L_2 with salts $CuCl_2 \cdot 2H_2O$ and $CoCl_2 \cdot 6H_2O$ and L_4 with salts $CrCl_3 \cdot 6HO$ and $CdCl_2 \cdot H_2O$. The analytical procedures for CPE which were designed for the determination of analytes including optimization of the Parameters for CPE including (reagent concentration, HCl concentration, incubation time, equilibration temperature, thermodynamic study and Triton X-100 volume) were selected in this study. The present CPE method was applied for determination concentration of metal ions (Cd^{2+} , Cu^{2+} and Co^{2+}) in real samples by FAAS.

Chapter Three included the new azo-azomethine compounds prepared and their complexes which were characterized by various analytical techniques as a UV-Visible spectroscopy(UV-Vis), Infrared Spectroscopy(FT-IR), Nuclear Magnetic Resonance Spectroscopy(^1H -NMR), Mass Spectra and Molar Electrical Conductivity for complexes and the results were identical to what is expected scientifically. The spatial shape of the complexes which was prepared is (octahedral) and stoichiometry of the complex. It's ratio 1:1.

In chapter three, there are too highlights on the results and discussion in analytical applications for the separation and preconcentration for the metal with separation and extraction method, such as cloud-point extraction (CPE) as an separation technique that uses a surfactant, instead of organic solvents for phase separation.

In the first part of CPE, Analytical reagent namely *6-((1E)-((2-(((E)-2-hydroxy-3-methoxy-4-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)benzylidene)amino)benzyl)imino)methyl)-2-methoxy-3- ((6-methoxybenzo[d]thiazol-2-yl)diazenyl)phenol (L_1)* after synthesis and characterization by using various analytical techniques as a preliminary

step toward it's using as an organic reagent for the extraction process involve the conversion of cadmium(II) ion in acidic into chloro anion CdCl_4^{2-} complex and subsequently the hydrophobic ion-pair complex formed with (L_1) cation was extracted into the nonionic surfactant (Triton X-100) rich phase at a temperature above its cloud point (CPT). Separation of cadmium ion by CPE involves the preceding formation of complex with the sufficient hydrophobicity to be extract into the small volume of surfactant rich phase as CdCl_4^{2-} . Accordingly, two separation schemes were designed and studied with CPE for the determination of Cd(II) as CdCl_4^{2-} using cloud point extraction (CPE) methodology by FAAS and the understanding of the mechanism of solubilisation and phases separation via the extensive thermodynamic studies of these forms in CPE system. Various parameters that can impact the extraction efficiency of these species are optimized by using one-factor-at-a-time (OFAT) procedure such as organic reagent concentration, HCl concentration, incubation time, equilibration temperature and Triton X-100 amount. Under its optimum conditions, the obtained results showed the enrichment factor of 51.259 which is achieved for Cd with the L_1 . The linear range for Cd^{2+} of $(0.01-1.3) \mu\text{g}.\text{ml}^{-1}$ is with detection limit of $0.012464 \mu\text{g ml}^{-1}$. The precision (%RSD, $n=8$) of the proposed is %1.308 at $0.05 \mu\text{g ml}^{-1}$ Cd (II), the developed method was applied for the determination of Cd(II) concentration in real samples including river, tap waters, milk, tea, tobacco and soil. The accuracy was determined by recovery percentage for real samples, it's found good recoveries (100.165-103.383%) for Cd^{2+} with L_1 .

In the second part of CPE, Analytical reagent namely **6,6'-((1E,1'E)-(1,2-phenylenebis(azanylylidene))bis (methanylylidene))bis(3-(phenyldiazenyl)phenol)(L_2)** was synthesised and fully characterized by

using various analytical techniques as a preliminary step toward its using as an organic reagent for the formation of an ion-association complex with copper ion in acidic medium, apt to interact with surfactant in solution. The L_2 as a reagent for the formation of an ion association complex with copper in acidic solution and the complex is extracted into the surfactant Triton X-100 at optimum conditions. The surfactant-rich phase which contains copper complex is mediated with solvent and the Cu content measured by FAAS ($\lambda_{\max} = 324.75 \text{ nm}$). The effects of the several variables which affect the CPE efficiency are optimized by OFAT procedure beside other parameters with which one is mentioned with Cu(II). The thermodynamic study was also investigated to expand the understanding of the mechanism of solvation of Cu(II) in the micelles. Under the optimized condition established, the enrichment factor of 52.574 was achieved for Cu^{2+} with the L_2 . The concentration range of $(0.01\text{-}3) \mu\text{g ml}^{-1}$ leads to detection limit of $0.017238 \mu\text{g ml}^{-1}$. The precision for ((%RSD, $n=8$) at $0.05 \mu\text{g.ml}^{-1} \text{ Cu}^{2+}$ was of %1.063. The developed method was used for the determination concentration of Cu^{2+} in real samples. The accuracy was determined by recovery percentage for real samples, it's found good recoveries (100.380-103.377%) for Cu^{2+} with L_2 .

In the third part CPE, Analytical reagent namely **6-(((2-(((E)-2-hydroxy-3-methoxy-4-(o-tolyldiazenyl)benzylidene)amino)benzyl)imino)methyl)-2-methoxy-3-(o-tolyldiazenyl)phenol(L_3)** was synthesised and fully characterized by using various analytical techniques as a preliminary step toward its using as an organic reagent for the formation of an ion-association complex with cobalt ion in acidic medium and the complex is extracted into the surfactant Triton X-100 at optimum conditions for determination of Co(II) as CoCl_4^{2-} ion by using the combined cloud point extraction (CPE) methodology. After phase

separation, the surfactant-rich phase was diluted with 1M HNO₃ in methanol and cobalt (II) ion determined by FAAS technique at λ_{max} of 240.72 nm. The optimization of complexation and extraction conditions was investigated. Thermodynamic parameters of CPE for solubilization process of the ion-pair complex in Triton X-100 were also considered. Under the optimized conditions, the preconcentration of a 15ml sample which gave preconcentration and enrichment factors were of 15 and 54.078 respectively. The calibration graph was linear in the range of (0.025-3) $\mu\text{g ml}^{-1}$ with a limit of detection and quantitation limit 0.011534 and 0.038446 $\mu\text{g ml}^{-1}$. The relative standard deviation for replicate determinations at 0.05 $\mu\text{g ml}^{-1}$ level was of (0.823%, n=8). The proposed method was applied for the determination of cobalt in river, tap waters, milk, black tea , tobacco and soil by FAAS. The accuracy was determined by recovery percentage for real samples, it's found good recoveries (101.108-104.918%) for Co²⁺ with L₃ .

The study of the stoichiometry for ion association complexes using the slope analysis technique showed that the ion association complex ratio was 1:1 for all extracted complexes

Chapter One

Introduction

Introduction

1.1: Azo - Azomethine dyes

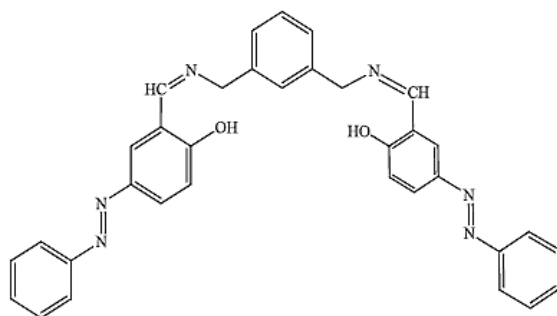
Azo and azomethine dyes are organic dyes that contain the characteristic --N=N-- and --CH=N-- chromophores⁽¹⁾. Azo-azomethine compounds are derived from the reaction between the Schiff base and azo represented in important series of widely studies organic ligands⁽²⁾. Azo-Schiff bases, having imine groups(C=N), are formed by the condensation reaction of (NH_2) primary amines with (CO) aldehydes or ketones⁽³⁻⁵⁾ and those having azo groups (--N=N--), are formed by a coupling reaction between a diazonium salt and a coupling agent⁽⁶⁾. A large number of (N,N') - donor ligands in azo imine family have been prepared in the last few years^(7,8). The spectral properties of azo dyes depend on the nature of both the azo and the coupling components. For example, azomethine compounds which have some interesting structural properties and uses can be prepared by coupling of azo and methine groups^(9,10). Azo-azomethine dyes are relatively robust and chemically stable and their spectral properties depend on the combination of electron-donating and withdrawing moieties in the molecules⁽¹¹⁾. Furthermore, considerable attentions are paid to the study of azo-azomethine dyes containing hydroxyl groups due to their metabolites which are toxic and carcinogenic^(12,13). The prototropic properties of hydroxyl groups of those dyes have important consequences on their electronic structure which can be exploited for their thermochromic and/or photochromic behavior⁽¹⁴⁾ and also for the design of molecular electronic devices⁽¹⁵⁾. However, photo-physical and photo-chemical properties of polyhydroxy azo-azomethine dyes are in turn strongly influenced by several factors including temperature, substituent groups and solvent polarity^(16,17).

This class of azo compounds possess active (π - acidic) azo imine ($-N=N-$, $CH=N-$) function as efficient agents to stabilize low valent metal oxidation states due to the presence of azo-centered π^* -molecular orbital, for this reason a number of these compounds were prepared and their abilities as chelating ligands were investigated⁽¹⁸⁻²⁰⁾. All of imine, azo and imine-azo product derivatives contain chromophoric group. They will examine the change in the color of solution through the addition of base and acid. Azo group indicates stabilization in the acid and base condition while the imine could be hydrolyzed in the base condition⁽²¹⁾. p-Acceptor ligands such as azo-containing compounds or an imines are capable of stabilizing metals in formally low oxidation states⁽²²⁾. Schiff bases are derived from azo chromophore group containing aromatic aldehydes have been extensively studied⁽²³⁾. Azo-azomethine ligands contain both azo and azomethine groups. Azo groups possess excellent donor properties and play an important role in coordination chemistry^(23,24). These compounds have received special attention in coordination chemistry due to their mixed hard-soft donor character and versatile coordination behavior⁽²⁵⁾. Azo-azomethines are known to be interesting because of the existence of both hard nitrogen and/or oxygen donor atoms in the backbones of these compounds, some of which have interesting physical and chemical properties⁽²⁶⁾. Therefore, several studies have been published on the synthesis and spectral properties of azo-azomethine dyes, as well as their transition metal complexes⁽²⁷⁻²⁹⁾. All colors of dye are available in azo dyestuff: yellow, red, purple, blue, green, brown, and black) as the amount of azo-groups increases, the intensity of the color increases. In addition, the color intensity increases when groups such as - OH, - NH₂, - N(CH₃)₂, - CH₃ and halogens are abundant and when

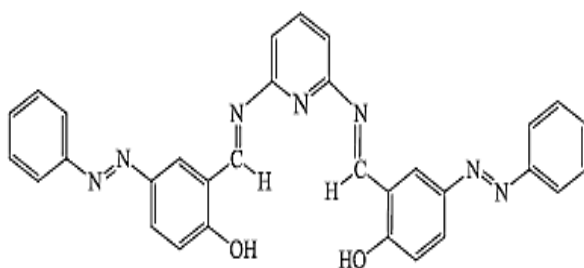
naphthalene derivatives, instead of benzene derivatives, are used⁽³⁰⁾. Azo-azomethine dyes have the ability to form different type of intra- and intermolecular hydrogen bond and intramolecular proton transfer between their nitrogen atoms. This tautomerization can be induced either by light, heat or the solvent^(31,32). Intermolecular proton transfer of this class of dyes has important consequences on their electronic structure which may be exploited for their photochromic and/or thermochromics behavior⁽³³⁾. Azo colorants and salicylaldehyde azo are a part of our everyday colorful life, thus, the study of the relationship between their structure and optical behaviors in different media will be most interesting for practical applications. Furthermore, the substituent change in salicylaldehyde azo and related schiff bases alters the photophysical properties such as electronic structure, ground and excited state dipole moments and intra- and inter-molecular interactions in solutions. Along with substitution changes, solvent effects can play a significant role in many chemical and physical behaviors such as hydrogen bonding and tautomerism of Schiff bases in solutions⁽³⁴⁾. Therefore, it is remarkable that azomethine containing azo dyes are forming complexes with certain metals. Obtained metal complexes are used as nonlinear optic materials. The crucial point of nonlinear optic material synthesis, is the highly polarizable characteristic of the molecule to be synthesized. To provide this, substituent groups of different characteristics must be conjugated to the structure⁽³⁵⁾. These groups are compelled to be good electron donors and acceptors, so that the polarizability of the molecule increases⁽¹¹⁾. Azo-schiff bases play an important role in inorganic chemistry as they easily form stable complexes with most transition metal ions such as cobalt(II), nickel(II) and copper(II). The development of the field of bioinorganic

chemistry has increased the interest in Schiff base complexes, since it is recognized that many of these complexes may serve as models for biologically important species⁽³⁶⁾.

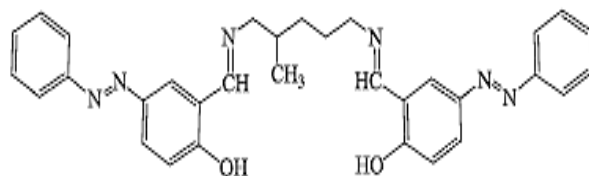
Because of the importance of azo-containing Schiff base compounds and in continuance of the interest in syntheses of azo-schiff base compounds, Eissa⁽³⁷⁾ was prepared of new Azo schiff bases by condensation of 3-formyl-4-hydroxy phenylazo benzene. With both 1,3-diamino xylylene, 2,6-diamino Pyridine, 1,5-diamino-2-methyl pentane respectively in (scheme1). The (I,II,III) compounds are new and prepared for the first time. The new compounds were identified by ¹H-NMR, IR, LC-MS spectral methods and were studied for antibacterial activities against (*Bacillus subtilis* and *Staphylococcus aureus*) which are gram positive and (*Salmonella typhi* and *Esche-richia coli*) that are gram negative. The results show that their activities were found to vary from moderate to very strong .



Compound I



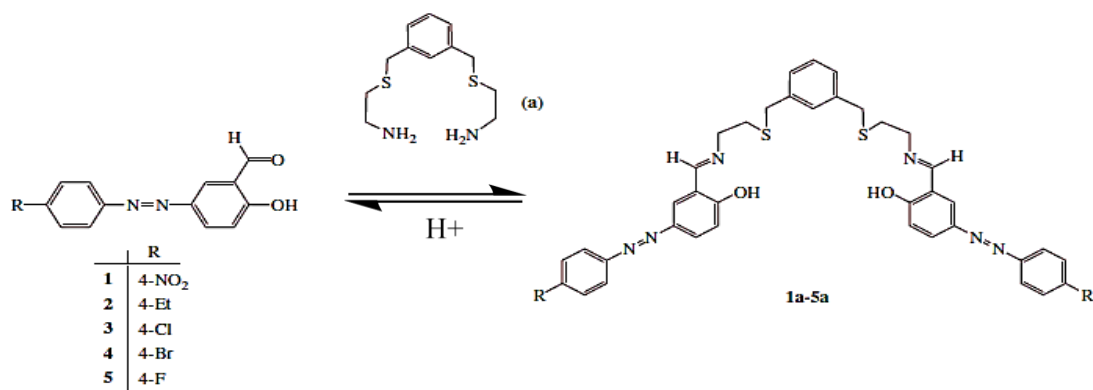
Compound II



Compound III

Scheme 1: Azo-Schiff base compounds

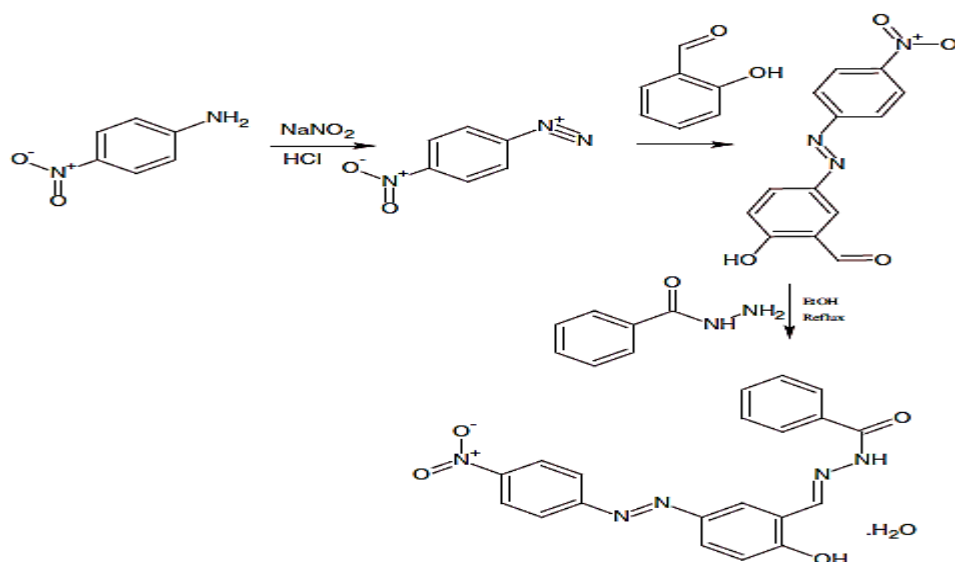
Khanmohammadi H. and Khodam F.⁽³⁸⁾ were synthesized of a new series of azo-azomethine dyes with $N_2S_2O_2$ donor set of atoms which were synthesized via condensation reaction of α,α' -bis(2-aminoethylthio)-1,3-xylene and substituted azo-coupled salicylaldehyde in (scheme 2). The dyes were characterized by IR, UV-Vis and ^1H - NMR spectroscopic methods as well as elemental analysis.



Scheme 2: New series of azo-azomethine dyes

Bitmez S.et.al⁽³⁹⁾ were synthesized of a new tridentate azo-azomethine ligand, N'-[2-hydroxy-5-[(4-nitrophenyl)diazenyl]phenyl]methylidene] Benzohydrazidemonohydrate,(sbH.H₂O), which is prepared by condensation of benzohydrazide and 2-hydroxy-5-[(4-nitrophenyl) diazenyl]benzaldehyde from treatment of a solution of diazonium salt of p-nitroaniline and 2- hydroxybenzaldehyde in EtOH which is shown in (scheme 3). The structures of the compounds are elucidated from the elemental analyses data and spectroscopic studies. It is found that the

ligand acts as a tridentate bending through phenolic and carbonyl oxygens and nitrogen atom of the C=N-group similar to the most of salicylaldehydes.



Scheme 3: Synthesis of azo-azomethine (sbH.H₂O)

Azo schiff base dyes are derived from the reaction between the schiff base and azo compounds .

1.2: Azo compounds

Azo dyes are chemical compounds that have the R-N=N-R' functional group, where R and R' can either be alkyl or aryl group⁽⁴⁰⁾. Azo dyes are derivatives of diimide (also called diazene), HN=NH , in which the two hydrogen atoms can be replaced with aliphatic or aromatic groups⁽⁴¹⁾. The name azo refers to the N=N and is taken from the French name for nitrogen, which is azote. The original word itself is from Greek meaning without “a” life “zoe”⁽⁴²⁾. Azo dyes can be classified according to the number of azo groups that is found in the compound: monoazo for one, diazo for two, or triazo for three azo groups. They can also be classified according to the auxochromic group that is found in it, for

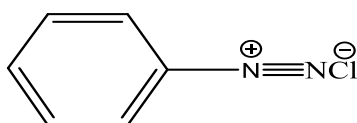
example, acidic azo dyes (that have acidic groups such as $-\text{SO}_3\text{H}$, $-\text{COOH}$) and basic dyes (that have basic groups such as dialkylamino, alkylamino and amino)⁽⁴³⁾. Approximately 70-80% of all dyes in textile manufacture and food industry are azo dyes. Although azo dyes can provide all the colors of the visible spectrum, in practice the yellow/red dyes are more prominent than the blue/brown dyes⁽⁴⁴⁾. Azo compounds are very important chemical compounds⁽⁴⁵⁾, receiving attention in many areas of research. They are colored, used as dyes and pigments for a long time⁽⁴⁶⁾. Azo dyes are easily prepared using the coupling reactions⁽⁴⁷⁾. Azo-coupling is one of the most important reactions for combining aromatic rings⁽⁴⁸⁾ and preparing azobenzene derivatives which contain active functional groups as a precursor for further synthesis to give different organic molecules such as : azo-amide⁽⁴⁹⁾, azo-imine⁽⁵⁰⁾.

1.3: Synthesis of Azo dyes

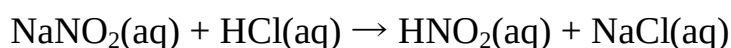
The general synthetic route of azo compounds involves two steps: diazotization and azo coupling.

1.3.1: Diazotization

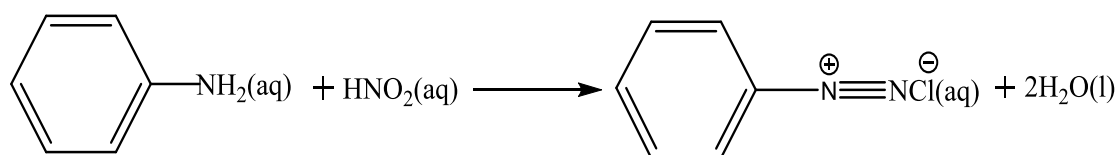
The diazonium salts are very unstable; the only relatively stable diazonium salts are the aromatic ones, and these are not particularly stable. This is because the presence of the benzene ring with its high electron density stabilises the $\text{N}\equiv\text{N}$ group. Benzenediazonium chloride is an example of a diazonium salt :



in aqueous solution, Benzenediazonium chloride decomposes above temperatures of 5°C and the solid compound is explosive; for this reason, diazonium salts are prepared in ice-cold solutions and are used immediately. They are synthesised in a diazotisation reaction a cold solution of sodium nitrate which is added to a solution of arylamine in concentrated acid (below 5 °C). The acid firstly reacts with the sodium nitrate to form an unstable nitrous acid⁽⁵¹⁾ :



The nitrous acid then reacts with the arylamine:

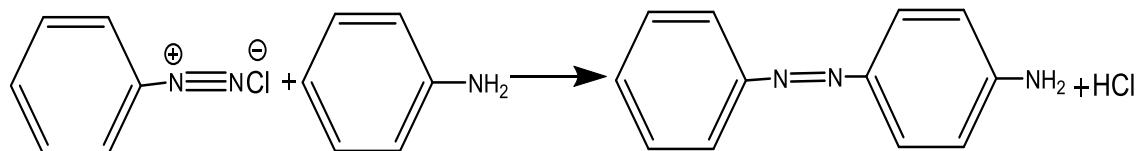


1.3.2: Diazo Coupling Reactions

In a diazo coupling reaction, the diazonium salt reacts with another arene (the coupling agent) . The diazonium salt acts as an electrophile, reacting with the benzene ring of the coupling agent. When the ice-cold solution of the diazonium salt is added to a solution containing the coupling agent, a coloured precipitate of an azo compound is formed, many of these compounds are dyes. The coupling agent always reacts in the ortho or para positions of the benzene ring (where one position is the functional group). The colour of the compound formed depends on the coupling agent that is being reacted with diazonium salt⁽⁵²⁾ .

1.3.2.1: Coupling with Amines

The coupling is often formed when a diazonium salt is reacted with arylamines .

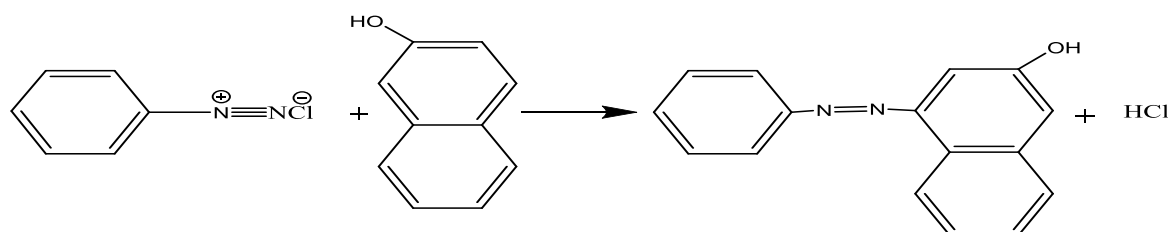


Scheme 4: Coupling with amine

Many different azo compounds can be formed by coupling different diazonium salts .

1.3.2.2: Coupling with *B*-Naphthols

The coupling is often when a diazonium salt is reacted with a *B*-naphthol⁽⁵³⁾



Scheme 5: Coupling with *B*-naphthol

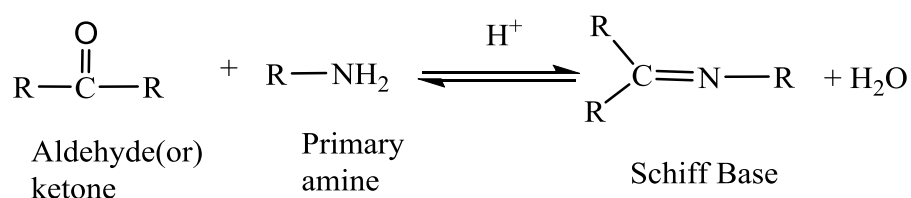
1.4: Schiff base ligands

1.4.1: Historical background of schiff base ligands

Schiff base ligands are also known as imines or azomethine that were discovered by Hugo Schiff, a German scientist after condensing primary amines with carbonyl compounds in 1864⁽⁵⁴⁾. The schiff base ligands are compounds that contain carbon-nitrogen double bond backbone $\text{R}_3\text{R}_2\text{C}=\text{NR}_1$ ⁽⁵⁴⁾. The substituents R_1 , R_2 and R_3 may be alkyl, aryl, heteroaryl or hydrogen. Schiff base compound was found to be useful as ligands for preparation of metal complexes and as bioactive agents in medicinal fields⁽⁵⁵⁾.

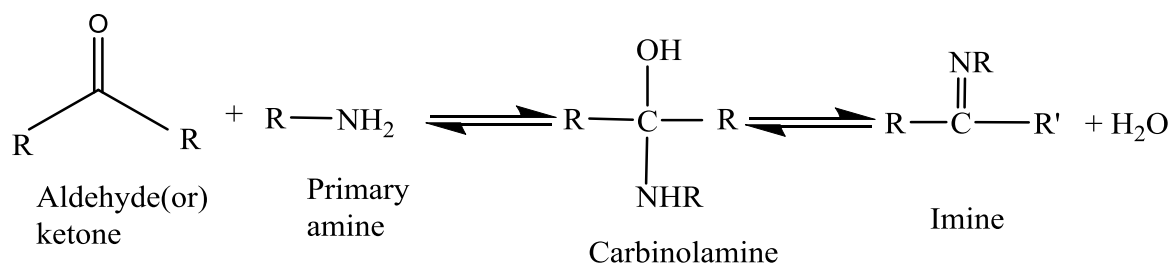
1.4.2: The chemistry of Schiff base ligands

The condensation of primary amines with aldehydes or ketones gives imines. Imines that contain an aryl group bound to the nitrogen or to the carbon atom are called schiff bases⁽⁵⁶⁾.



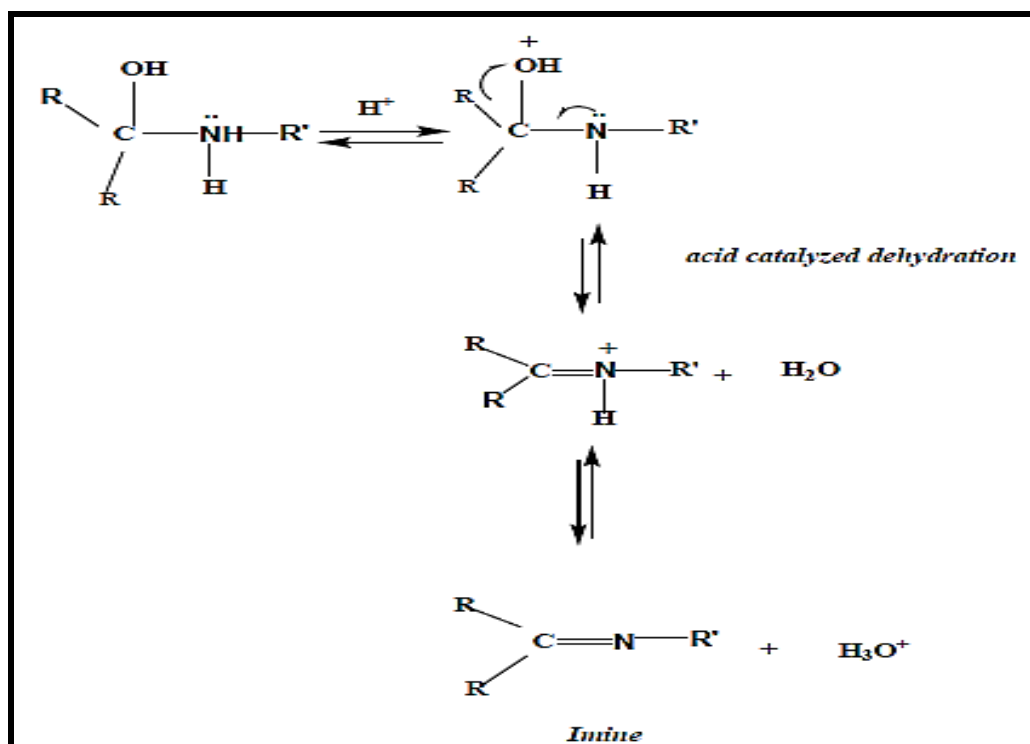
Scheme 6: Formation of schiff base

The formation of a schiff base from an aldehydes or ketones is a reversible reaction and generally takes place under acid or base catalysis, or upon heating. The formation is generally derived to the completion by the separation of the product or the removal of water, or both. Many schiff bases can be hydrolyzed back to their aldehydes or ketones and amines by acid or base^(57,58). The mechanism of schiff base formation is including of neucleophilic addition to the carbonyl group. In this case, the neucleophile is the amine. In the first part of the mechanism, the amine reacts with the aldehyde or ketone to give an unstable addition compound called carbinolamine.



Scheme 7: Formation of Schiff base with carbinolamine intermediate

The carbinolamine loses water by either acid or base catalyzed pathways. Since the carbinolamine is an alcohol, it undergoes acid catalyzed dehydration. The dehydration of carbinolamine is also catalyzed by base. This reaction is somewhat analogous to the E₂ elimination of alkyl halides. The Schiff base formation is really a sequence of two types of reactions, i.e. addition followed by elimination. Yet the acid concentration cannot be too high because amines are basic compounds. If the amine is protonated and becomes non-nucleophile, equilibrium will be pulled to the left and carbinolamine formation cannot occur. Therefore, many schiff bases synthesis are best carried out at mild acidic⁽⁵⁹⁾.



Scheme 8 : The mechanism of Schiff base under acidic conditions

Schiff base ligands are relatively stable, however, owing to the imine group in these compounds they are susceptible to hydrolysis and transamination by nucleophiles⁽⁶⁰⁾. Schiff base ligands have the ability to coordinate as mono-dentate to multi-dentate such as tetra-dentate, tri-dentate, bi-dentate and chelation abilities in the presence of donor sites⁽⁶¹⁾.

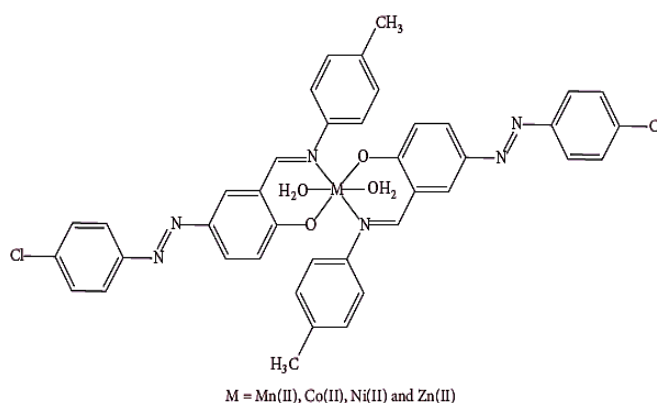
1.5: Azo-Azomethine complexes

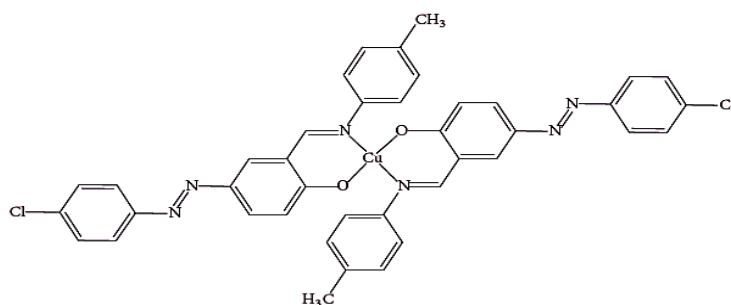
Azo and azomethine dyes are organic dyes that contain the characteristic --N=N-- and --CH=N-- chromophores. Metal complexes of these dyes are considered to be important stereochemical models in transition metal coordination chemistry due to their preparative accessibility and structural variety⁽¹⁾. The properties of schiff base ligand varies based on substituents bonded to the imine functional group. The coordination of metals to schiff base ligands takes place through the imine nitrogen and another group, which is connected to the aldehyde used to form the ligand⁽⁶²⁾. Azo group is characterized by a lone pair of orbital containing two electrons on nitrogen atom, if linked to an aromatic ring carrying an additional donor sites is well suitable for chelation and its complexes contain both azo and azomethine groups⁽⁶³⁾. Azo-azomethine metal complexes have been studied extensively for years due to the synthetic flexibilities of these schiff base ligands and their selectivity as well as sensitivity towards the transition metal ions⁽⁶⁴⁾.

These compounds have a great ability to coordinate with many metal ions and form stable complexes. The coordination compounds of azo-azomethine ligands are also widely used in medicine, for corrosion prevention⁽⁶⁵⁾. Some azo containing schiff base complexes were also used as catalysts in many organic reactions such as oxidations or

reductions on many chemical compounds⁽⁶⁶⁾. In addition to the above features, the catalysis of alkene oxidation by soluble transition metal complexes is of great interest in both biomimetic and synthetic chemistry⁽⁶⁷⁾. Because the oxidation reactions are carried out in elevated temperatures, it has become important to synthesize thermally stable schiff base complexes as catalysts⁽⁶⁸⁾. Azo schiff base and their complexes with transition metal ions are also of importance due to their complexing, catalytical, biological properties⁽⁶⁹⁾.

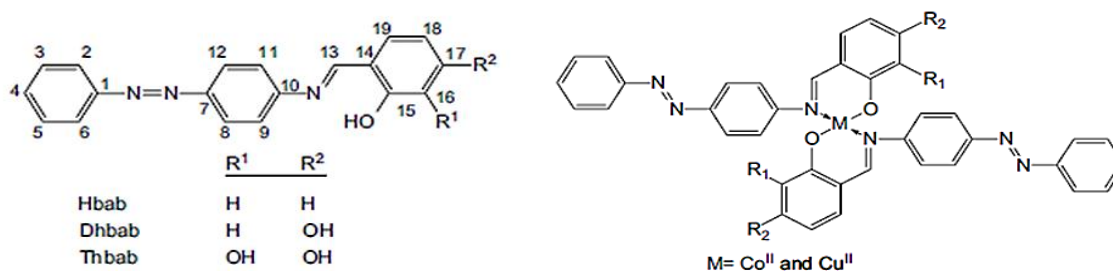
Anitha C. *et.al*⁽⁷⁰⁾ were synthesized and characterized of azo schiff base complexes of Co(II), Ni(II), Cu(II), Mn(II), and Zn(II) from 4-((4-chlorophenyl)diazenyl)-2-((*p*-tolylimino)methyl) phenol (CDTMP) in (scheme 9). The nature of bonding and the structural features of the complexes have been deduced from elemental analysis, molar conductance measurements, IR, UV-Vis, ¹H-NMR and mass spectra. Spectroscopic and other analytical studies reveal square-planar geometry for copper and octahedral geometry for other complexes and the obtained data showed that the stoichiometry of the chelates is 1: 2 (M:L) and the analytical data of the complexes are in good agreement with the general formula ML₂.





Scheme 9: Synthesis of azo-schiff base complexes

Ispir E.⁽⁹⁾ was synthesized and characterized of a series of azo-linked salicylidenic schiff bases named 2-((E)-(4-((E)-phenyldiazenyl) phenyl imino)methyl)phenol (Hbab), 4-((E)-(4-((E)-phenyldiazenyl) phenyl imino)methyl)benzene-1,3-diol (Dhbab) and 4-((E)-(4-((E)-phenyldiazenyl) phenyl imino)methyl)benzene-1,2,3-triol (Thbab) and their Co(II) and Cu(II) complexes in (scheme 10). The ligands were synthesized by the reaction of p-aminoazobenzene with salicylaldehyde, 2,4-dihydroxybenzaldehyde and 2,3,4-trihydroxybenzaldehyde, respectively. All prepared compounds were characterized using elemental analyses, IR, UV-visible spectroscopy and conductance measurements. According to the analytical, UV-visible and IR data, the Co(II) and Cu(II) metal complexes are formed by the coordination of N, O atoms of the ligands. The analytical data shows that the metal to ligand ratio in the mononuclear schiff base complexes is 1:2.



Scheme 10: Synthesis of series of azo-linked salicylidenic schiff bases

1.6: Extraction Techniques

In chemical analysis separation process including the separation of a substance from a matrix. In spite of the development and advancement of modern analytical instrumentation, direct analysis of ultra-trace amount of compounds and metals in complex matrix is still difficult and sometimes it is an impossible task than the determination of total compounds contents because:-

For compounds present at ultra- trace amount, few suitable analytical procedures.

There are difficulties associated with isolating the targeting compound from complex matrices.

Most of the techniques available disturb the equilibrium existing between the various compounds which are present in the system under study and suitable standard reference materials are of the unavailable⁽⁷¹⁾.

In such matrices, the detection be supposed to preceded by separation and preconcentration simultaneously. This provide lowering the detection limit, minimizing contaminants, and eliminating matrix effect of many compounds with different techniques. Preconcentration techniques ought to be touchy, and particular for smaller scale segments, speed, simple procedure among others, and ecologically safe. There are several separation/ preconcentration methods reported in the chemical literature and among them such as liquid-liquid extraction (LLE)⁽⁷²⁾ and solid phase extraction (SPE), but each has its own disadvantages. For example, LLE used large amounts of organic solvents which make this method harmful to the analysts and environment because of its toxicity (i.e. eco-unfriendly), labor to be intensive, expensive and time-

consuming. Whilst SPE used less organic solvents but it is not available in each laboratory, more expensive and when it be automated entails additional cost and complexity⁽⁷³⁾.

Therefore, there is a great need for new separation/ preconcentration method to reach to the modern analytical methods. In this respect, several separation / preconcentration techniques came into being that using and consumption of minimum solvents and reduce in the laboratory wastes. These methods include, solid-phase micro extraction (SPME), liquid phase micro extraction (LPME), homogenous liquid liquid extraction (HLLLE), dispersive liquid–liquid micro extraction (DLLME), single drop micro extraction (SDME) and cloud-point extraction (CPE)⁽⁷⁴⁾.

Despite SPE and its counterpart methods like SPME, LPME and DLLME are complied with green chemistry criteria and their success in the separation and enrichment of diverse analytes in different samples matrices due to their ability for automation with analytical instruments. But they are not without disadvantages, such as those matrix interferences, using slightly more organic toxic solvent and forming secondary waste, some of them are restricted in using with specific instruments, poor enhancement factors, relatively expensive, time-consuming and unavailable in many laboratories⁽⁷⁵⁾.

Cloud point extraction has two directions of benefits, the first is the selective extraction because of using a special type of surfactant, and the second is the preconcentration for the analyte before determination in instrument. The comparison between the extraction techniques is summarized in table (1.1) .

Table (1.1): Comparison of different extraction techniques for liquid samples⁽⁷⁶⁾

Parameter	LLE	SPE	SPME	CPE
Brief description	Analyte is partitioned between two immiscible solvents	analyte retained on a solid adsorbent	Partition equilibrium of analytes between a polymeric stationary phase and the sample matrix	Analyte is partitioned between two isotropic phase “surfactant-rich phase” and “aqueous phase”
Extraction time	Up to 24 h	20–30 min	Up to 60 min	20 min
Solvent usage	Organic solvent	Organic solvent	None	Surfactant solution
Cost	Low cost	Relatively low cost	Relatively low cost	Low cost
Ease of operation	Easy	Relatively easy	Relatively easy	Easy
Disadvantages	Large consumption of solvent and concentration of sample required after extraction	Insufficient retention of very polar compounds and limited selectivity	Choice of fiber and optimization of procedure required	Optimization of operating conditions

1.7: Cloud-Point Extraction (CPE)

The potential of micellar systems as an alternative to organic solvent was extensively introduced in the extraction and preconcentration of metal ions and organic compounds and became one of the most preferred preconcentration steps for enhancing the sensitivity in metal and organic analysis and an important practical application in the use of surfactants in analytical chemistry⁽⁷⁷⁾. This methodology was named “cloud point extraction” and abbreviated as CPE or “micelle-mediated extraction” abbreviated as MME.

The first usage CPE comes back to the works of Watanabe and co-

workers⁽⁷⁸⁾ who recommended the application of CPE by employing the non-ionic surfactants as an alternative solvent for the extraction of metal ions^(77,79). In principle, CPE depends on the separation of hydrophobic analyte (i.e. inorganic and/or organic analyte) between two phases generated from micellar or non-ionic surfactant solution when heated to its critical cloud point temperature-CPT(i.e. the temperature above which an aqueous solution of water-soluble nonionic surfactant becomes turbid owing to the decrease in the solubility of the surfactant in water). One phase is called a surfactant-rich phase (SRP) with a small volume containing the analyte trapped by organized micelles, and other phase is called surfactant-poor phase (SPP) or bulk aqueous phase containing a little surfactant molecules at critical micelle concentration (CMC)⁽⁸⁰⁾.

Since its birth, CPE has been developed considerably in two parallel directions, both in term of CPE methodology and the analytical detection systems that accompanied the determination of analytes. The advancement in CPE methods has allocated among using various types of surfactants, chelating agents, electrolytes, etc. in order to improve the optimum conditions of the analytical CPE procedure and scoring a desirable enrichment factor of the analytes besides thermodynamic studies to understand the solubilization mechanism of hydrophobic analyte in the micelles. Also, the development involves the combination of CPE with various instrumental analytical techniques to achieve the best figures of merit of the analyte to be determined^(81,82). It is impossible to understand the action of micelles in a given analytical method or to accurately predict the results of incorporating micelles into a

new method of analysis without understanding of micelles themselves. This includes effects arising from molecular structure of amphiphile from which the micelle is derived and possible effect of the various substrates which might be present in the solution. Furthermore, the catalytic abilities of micelles must be recognized by the analyst as so much of the observed benefits of the use of micelles in analytical chemistry come from these abilities. There are many other properties of micelles which would be of some importance to researchers desiring to use them in analytical methods⁽⁸³⁾. Surfactants are amphiphilic molecules that are mean containing together hydrophobic group (tails) which is regularly extended chain of hydrocarbon or aromatic ring operates to reduce solubility in water, along with hydrophilic groups (heads) due to the occurrence of a number of nucleophilic or electrophilic groups which is often ionizable as well as has the differing effect of tail⁽⁸⁴⁾. Figure (1.1) viewing a plain structure of surfactant tiny part⁽⁸⁵⁾.

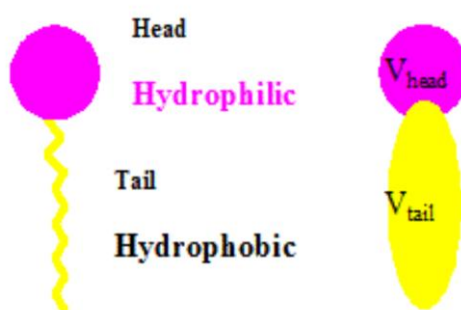


Figure (1.1): Simple structure of surfactant molecule

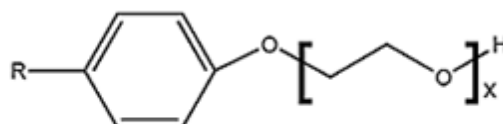
1.7.1: Selection and Properties of Surfactant

To date, the majority of the applications of CPE for metal and organic analysis focused on employing non-ionic surfactants type mainly, polyoxyethylenated alkylphenols ranging from the triton family to PONPE series. This is because they are all commercially available of high purity grade, low cost, stable, non-volatile, non-toxic, balanced hydrophobicity and environmentally friendly⁽⁸⁶⁾. Surfactants belong to a group of association or amphiphilic organic substances. Amphiphilic molecules consist of an ionic or nonionic polar head group and a hydrophobic portion. The formation of micelles or surfactant aggregates is primarily operated by three forces: the hydrophobic repulsion between the hydrocarbon chains and aqueous solution, the charge repulsion of ionic head groups, and the van der Waals attraction between the hydrocarbon tails⁽⁸⁷⁾. There are four types of general surfactants that are classified according to their charge on head group; non-ionic, cationic, anionic, and amphoteric (or zwitterions)⁽⁸⁸⁾. A large proportion of these nonionic surfactants are made hydrophilic by the presence of a polyethylene glycol chain, obtained by the polycondensation of ethylene oxide and called polyethoxylated (POE or R-OCH₂CH₂O-) nonionics . The cloud-point temperature depends on the structure of the surfactant and on its concentration. For ethoxylated nonionics which shows a peculiar behaviour with regard to temperature in a given temperature range, their solubility decreases with a temperature increase leading to surfactant precipitation at the “cloud point”. This anomalous behaviour of ethoxylated nonionics as well as the possibility to fine-tune the hydrophilic-lipophilic balance (HLB) makes them especially useful for bringing complex surfactant compositions to the phase-inversion

temperature (PIT) conditions . These surfactants are often referred to as polymeric ethers since they all contain common hydrophilic moiety of molecules, polyethylene oxide or polyethylene glycol, which has the repeated $(\text{CH}_2\text{CH}_2\text{O})_n$ ether structure with n generally in a range of 10 to 100 units⁽⁸⁹⁾.

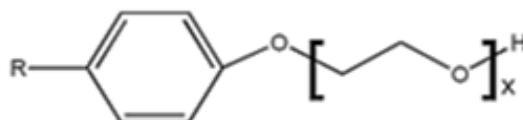
1.7.2: The Triton Family

The family is commercially available under different trademarks with a general chemical name as *tert*-octylphenol poly (ethyleneglycol ether)_n the slight differences between the products are in the average size (n) and the size distribution of the PEG-based head group. The classical phase is Triton X-100 ($n=9$ -9.6). The chemical name of Triton X-114 (or TX-114) is polyoxyethylene 7.5 octylphenyl ether and its structure is:



R = octyl (C8) TX - 114
X = 7.5 (avg)

It has a cloud point of 25 °C at 1% solution and its viscosity at 25°C is 260 cp⁽⁸⁸⁾ while the chemical name of Triton X-100(or TX-100) is a polyoxyethylene 9.5 octylphenyl ether and its structure is:



R = octyl (C8) TX-100
X = 9.5 (avg)

It has a cloud point of 64°C at 1% solution and its viscosity at 25°C is 240 cp⁽⁹⁰⁾.

1.7.3 : Separation Mechanism in CPE

In fact, the mechanism of separation for the CPE process is somewhat similar to that of conventional liquid-liquid extraction which separates the desired analytes by their affinity in two immiscible (homogeneous) liquid phases. But, in CPE process, the only difference being the “organic” phase (micelles) is generated within the aqueous phase, converting a previously homogeneous solution to heterogeneous one by simply gathering its previously scattered hydrophobic suspension⁽⁹¹⁾. CPE is a simple and straightforward extraction procedure (figure 1.2). A surfactant typically nonionic is added to an aqueous solution containing the species of interest. The concentration of the surfactant must be above the critical micelle concentration (CMC) of the surfactant which allows the surfactant to organize and form micelles. For metal ion extraction, a suitable chelating agent is added. In principle, the complexant should facilitate partitioning of the metal ion into the micelle by forming a hydrophobic complex with the metal. The properties of the aqueous solution are changed to reach the cloud point temperature (CPT), this can be done by addition of salt or other surfactants or with heating. Upon reaching the CPT, the solution becomes cloudy and eventually separates into two phases: the aqueous phase and the surfactant rich phase (SRP). The aqueous phase contains surfactant at a concentration less than or equal to the CMC, while the SRP contains the majority of the surfactant. The SRP is significantly smaller in volume than the aqueous phase, allowing for preconcentration of the target analyte.

The phase separation is reversible upon mixing and cooling below the CPT, these two phases merge to form a clear phase again⁽⁹²⁾.

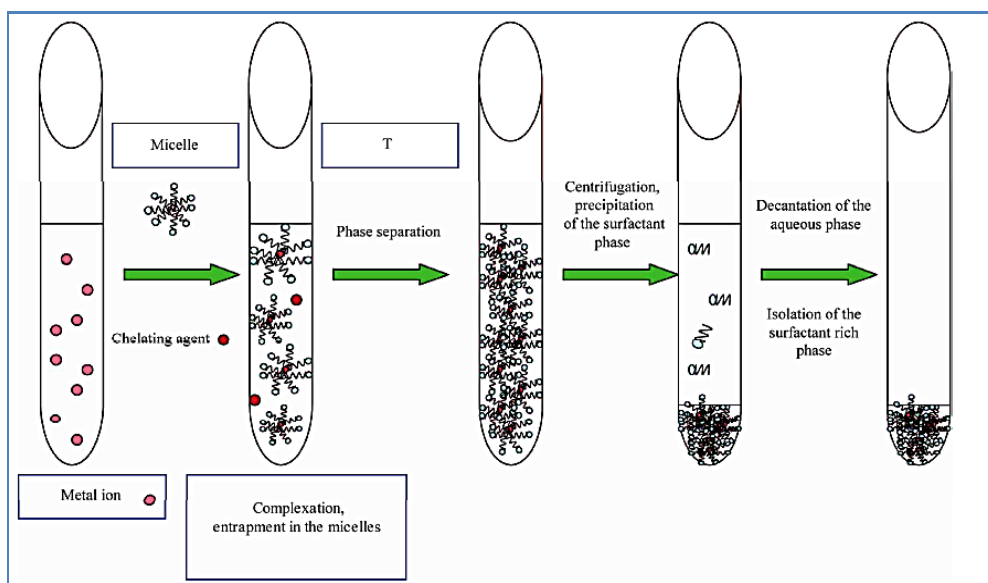


Figure (1.2): Separation mechanism in CPE process⁽⁹³⁾

1.8: Thermodynamic Approach of CPE

In fact, the clouding and separation mechanism phenomena in CPE are still to be defined exactly, several studies have shown that such phase separations result from the competition between entropy (which favours miscibility of micelles in water) and enthalpy (which favours separation)⁽⁹⁴⁾. An accurate explanation about cloud point phenomenon is still a controversy in the scientific literature. developed a model for explaining the phases separation in systems formed by non-ionic surfactants and water, at lower temperatures each surfactant's molecules are evolved by a highly organized water layer, which forms hydrogen bonds with the polar head groups and with the poly-oxyethylene units of non-ionic surfactants. When the temperature is increased, there is also an increase in the entropy, which causes the dehydration of polyoxyethylene

chains and destroys the water molecules layer. From this point, the weak Van der Waals forces among the molecules make important contributions to micellar agglomerate formation and consequentially, the phase separation occurs⁽⁹⁵⁾. The formation of aggregates of micelles at the cloud point is obviously guided by thermodynamic principles. At the phase separation temperature, the clouding components release their solvated water molecules and separate out from solution this can be considered as the limit of solubility. As phase separation is a reversible process, the chemical potential of species should be equal between surfactant-rich and surfactant-poor phases^(96,97).

1.9: General Analytical Procedure for CPE

CPE is a simple and straightforward extraction procedure and takes not more than half an hour to be accomplished after establishing the optimum conditions for the analyte determined. In fact, the experimental steps of CPE for metal and organic determination do not quite differ except in most cases the step of addition of chelating agent to metal ion to form hydrophobic complex to make easy extraction in the surfactant phase. But both producers engage in three simple main steps as follows; (1) solubilisation of the analytes in the micellar aggregates; (2) clouding; (3) phase separation for analysis⁽⁷⁹⁾.

In general, a typical CPE procedure for transition metal separation starts with an aqueous solution containing the metal. Ligand and surfactant are added to the aqueous solution in suitable quantities. Depending on the solubility of the ligand, it may be added pre-dissolved in the neat surfactant or to the aqueous solution. The solution is mixed to homogenize and then heated above the cloud point temperature to

promote phase separation. Once removed from heat, the sample is either placed on ice to increase the viscosity of the SRP or centrifuged to enhance phase separation. The SRP is typically on the bottom and the aqueous phase can be easily removed from the SRP for analysis. High performance liquid chromatography, flame atomic absorption spectroscopy and UV-visible spectroscopy have all been used to analyze in CPE⁽⁹⁸⁾. Due to its viscosity the SRP is difficult to analyze and must be diluted for analysis with ICP or HPLC⁽⁹⁸⁾. The general CPE procedure in metal analysis can be implemented via experimental steps as shown in figures (1.3) and (1.4).

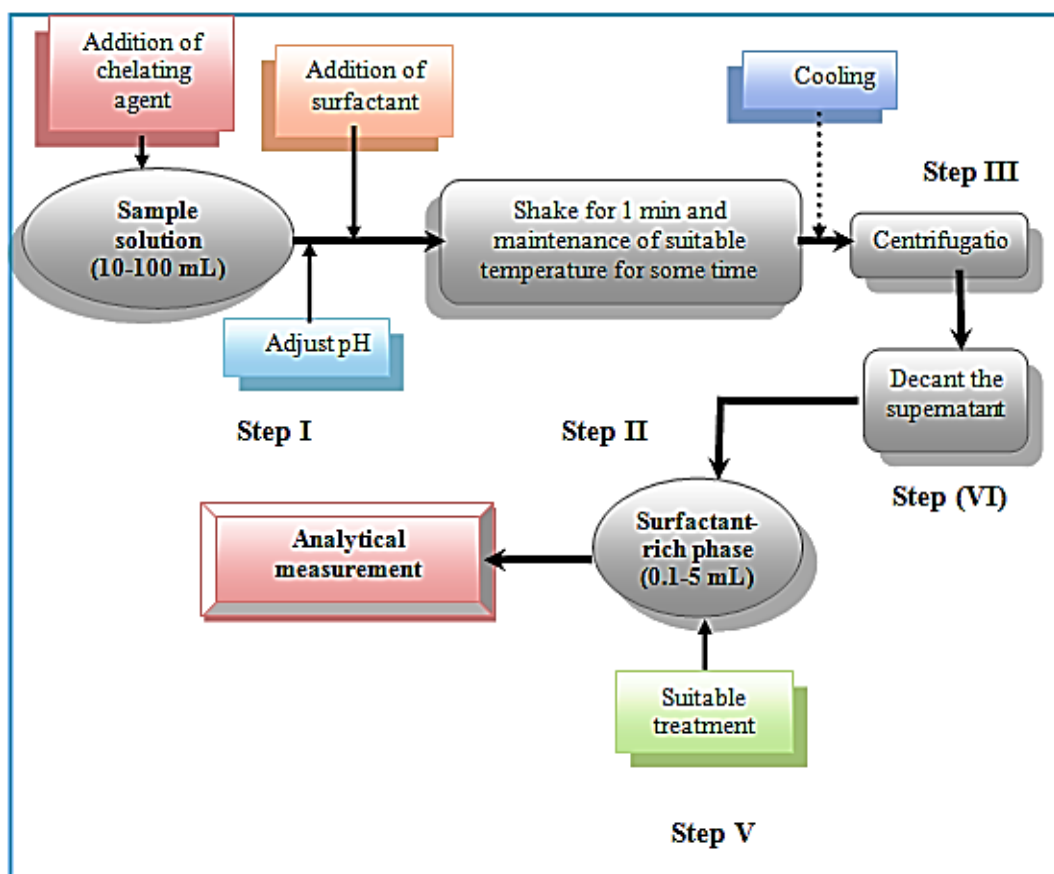


Figure (1.3): Flow chart of general systematic CPE procedure

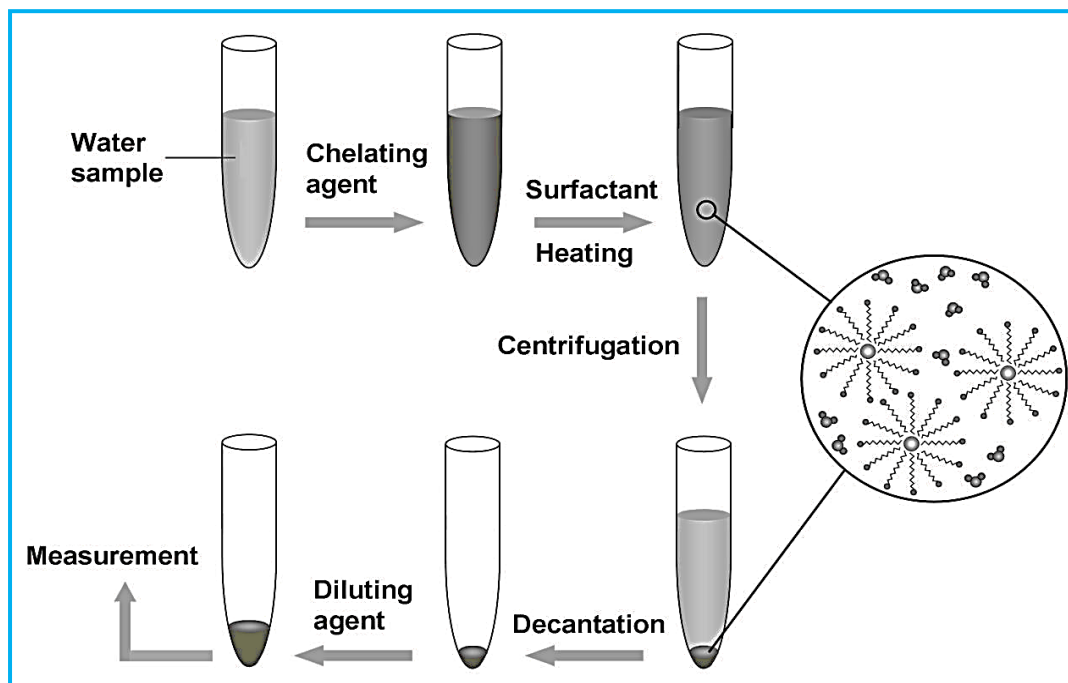
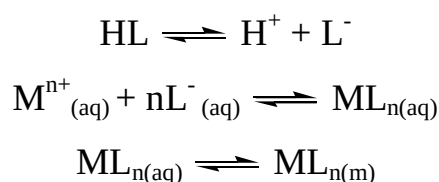


Figure (1.4): Schematic diagram of metal ion extraction in CPE procedure prior to its detection⁽⁹⁹⁾

The extraction process involves several equilibrium and chemical species. Simply, the acid dissociation step of chelating reagent and the formation of a hydrophobic chelate followed by its transference into a micellar phase can be represented by equations.



where $\text{M}^{n+}_{(\text{aq})}$ and $\text{L}^-_{(\text{aq})}$ are the metal and the ligand, respectively, in the aqueous phase, $\text{ML}_{n(\text{aq})}$ is the complex in the aqueous phase, and $\text{ML}_{n(\text{m})}$ is the complex in the surfactant phase. The distribution constant, K_d , the total formation constant, β_n , for the extracted species and the acid dissociation constant of HL, K_a , in aqueous phase is given by the following equations:

$$K_d = [ML_n]_m / [ML_n]_{aq} \quad \text{-----}(1)$$

$$\beta_n = [ML_n]_{(aq)} / [M^{n+}] [L^-]^n \quad \text{-----}(2)$$

$$K_a = [H^+] [L^-] / [HL] \quad \text{-----}(3)$$

Combining Eqs. (1)-(3) and rearranging gives:

$$[ML_n]_m / [ML_n]_{aq} = K_d \beta_n K_a^n [HL]^n [H^+]^{-n} \quad \text{-----}(4)$$

Since the distribution coefficient, D is given by:

$$D = \frac{(C)_m}{(C)_{aq}} = \frac{[ML_n]_m}{[M^{n+}]_{aq} + [ML_n]_{aq}} \quad \text{-----} (5)$$

assuming that $[M^n]_{aq} > [ML_n]_{aq}$, and combining Eqs. (4) and (5) results in:

$$D = K_d \beta_n K_a^n [HL]^n [H^+]^{-n} \quad \text{-----} (6)$$

Equation (6) shows that the distribution coefficient, D, is influenced by magnitudes of formation constant to specific cations with the complexing, by acid dissociation constant, the complexing reagent concentration, and the solution pH. These factors can be used to control the metallic ions extractions and must be evaluated to improve a method⁽⁸⁸⁾.

1.10: Heavy metals

The term Heavy Metals refer to any metallic element that has a relatively high density and it is toxic or poisonous at low concentration⁽¹⁰⁰⁾. Heavy metals are naturally occurring elements that have a high atomic weight and a density at least 5 times more than that of water⁽¹⁰¹⁾. Metals are vital for a huge number of physiological processes in the human body, But they can also destroy health when the concentration is not within the physiologically favorable range. Heavy

metals are important environmental pollutants and many of them are toxic even at very low concentrations⁽¹⁰²⁾. Even essential trace elements can cause damage to health or even death at increased concentrations. However, if the levels of these metals are higher than the levels of healthy life, the roles of these metals change to a negative dimension. The main sources of the heavy metal ions directly are food and water and, indirectly, industrial activities and traffic⁽¹⁰³⁾. The form in which an element is ingested also plays a major role in its toxicity⁽¹⁰⁴⁾. Metals can directly influence the behaviour by impairing mental and neurological function, influencing neurotransmitter production and utilization, and altering numerous metabolic body processes⁽¹⁰⁵⁾. Copper, cadmium and cobalt are metals with known toxic properties⁽¹⁰⁶⁾.

1.10.1: CPE for Cadmium Cd(II)

Cadmium is highly toxic even at very low concentrations causing damages to organs such as kidneys, liver and lungs⁽¹⁰⁷⁾. One of the pathways that cadmium enters human body is through daily intake of food and water, thus the monitoring of cadmium concentrations in food and water samples is of significant importance. The maximum contaminant level allowed by the American Environmental Protection Agency (US EPA) in standard drinking water is $10 \mu\text{g.l}^{-1}$ to provide ample protection of human health. Monitoring trace element concentrations in biological materials, particularly biological fluids, might be considered a difficult analytical task⁽¹⁰⁸⁾, mostly due to the complexity of the matrix and the low concentration of these elements, which requires sensitive instrumental techniques and often a preconcentration step. Concretely, spectrometric techniques for the

analysis of trace cadmium have developed rapidly due to the increasing need for accurate measurements at extremely low levels of this element in diverse matrices⁽¹⁰⁹⁾. The major route of cadmium exposure for the general population is via food. An increase in soil cadmium content generally results in an increase of plant uptake of cadmium although some soil and plant factors may influence cadmium accumulation by plants. Crops grown in cadmium contaminated areas have been found to contain elevated cadmium content compared with normal levels. Therefore, human cadmium exposure via food in contaminated areas can be many times above normal intakes and cadmium is known to be toxic for living and lead to cadmium toxicity organism even if it is present in low levels⁽¹¹⁰⁾. Some techniques extraction have offered proper extraction proficiency and high preconcentration factor. Be that as it may, they have introduced some of impediments related to test pollution, moderately tedious or high cost, unbending control of the conditions, require an extensive volume of natural solvents and generally poor precision. In this manner the separation and preconcentration of cadmium or other metal particles, after the arrangement of chelate, in view of cloud point extraction (CPE) as an option preconcentration strategy to elective extraction strategies, in the light of its effectiveness, effortlessness ease, industrially accessible surfactant, velocity and security⁽¹¹¹⁾. Countless manuscript have been available in the chemical literatures on the separation and preconcentration of cadmium in unusual matrices through

CPE methodology previous to its determination by means of different instrumental techniques as exposed in table (1.2).

Table(1.2): Cloud point extraction of Cadmium in different matrixes

Sample type	Chelating agent	Surfactant type	Detection system	PF	EF	LOD	Ref.
Tap water	IDPI	Triton X-114	FAAS	26	31	0.15 $\mu\text{g.ml}^{-1}$	12
soil, water and vegetables	CV	Triton X-100	Spectrophotometry	125	150	0.042 $\mu\text{g.ml}^{-1}$	113
Milk and cigarette	IPAI	Triton X-114	FAAS	30	33	2.4 ng.ml^{-1}	114
Natural water	PPT	Triton X-114	FAAS	100	-	0.38 $\mu\text{g.L}^{-1}$	115

1.10.2: CPE for Copper Cu(II)

Copper is an essential element not only for life in mammals but also for plants, and it plays an important role in carbohydrate and lipid metabolism. It has many biological effects as an essential element as well as a toxic one⁽¹¹⁶⁾. In general, a daily copper intake of 1.5-2 mg is essential for adults and copper at nearly 40 ng.ml^{-1} is required for normal metabolism of many living organisms^(116,117). Copper is a moderately toxic element as compared to other transition metals⁽¹¹⁸⁾. But, copper(II) at higher levels is toxic and severe oral intoxication will affect mainly the blood and kidneys. So, the trace copper(II) content in foods must be controlled on a daily basis and the European Commission has fixed the limit of 2 $\mu\text{g.ml}^{-1}$ for copper(II) in drinking water and the allowed limit of copper(II) is set to 1.3 $\mu\text{g.ml}^{-1}$ in the USA similar to that in Canada

(1.0 $\mu\text{g}.\text{ml}^{-1}$)⁽¹¹⁹⁾. Therefore, there is an increasing need to monitor copper(II) levels in food samples at ever decreasing concentrations. For this purpose, very sensitive, simple, rapid and inexpensive methods are necessary⁽¹²⁰⁾. Much interest and effort have been devoted on the studies for copper determination in water and biological matrices because it is a good tool for environmental and toxicological monitoring. The removal of copper from aqueous medium such as effluents also is of great interest for environmental and human health purposes⁽¹²¹⁾. The determination of trace copper in biological samples is particularly difficult because of the complex matrix and the usually low concentration of copper, which requires sensitive instrumental techniques and frequently a pre-concentration step⁽¹²²⁾. CPE separation, high enrichment factor, low consumption of organic solvents, detection techniques, much more environmentally friendly properties and it is safer because small volumes of noxious surfactants are used instead of toxic organic solvents⁽¹²³⁾. Separation and preconcentration based on CPE is becoming an important and practical application of surfactants in analytical chemistry. Table(1.3) compresses the uses of CPE for assurance of Cu(II) in various matrices.

Table (1.3): Cloud point extraction of Copper in different matrixes

Sample type	Chelating agent	Surfactant type	Detection system	EF	LOD	Ref.
environmental, water and biological samples	ANPAP	Triton X-114	Spectrophotometry	250	1.2 ng.ml ⁻¹	124
Tap, stream and rain water	SPAR	Triton X-100	FAAS	31	1.64 µg.L ⁻¹	125
milk powder and mineral water	DDTC	Triton X-114	FAAS	-	1.1 ng.ml ⁻¹	126
rice flour and water samples	MPMP	Triton X-114	FI-FAAS	81	15 µg.l ⁻¹	127

1.10.3: CPE for Cobalt Co(II)

Cobalt is an essential trace element which is present in most body tissues, with the highest concentrations typically found in the liver. It is a component of vitamin B12 (cyanocobalamin), which is involved in the production of the blood red cells and the prevention of pernicious anemia. Cobalt toxicity is quite low compared to many other metals. However, high exposure to this element can cause several health problems such as asthma and skin irritation. Therefore, quantitative determination of cobalt at low concentrations plays an important role in different fields such as environmental analysis, process control and medicine⁽¹²⁸⁾. Some of heavy metals like cobalt is necessary for human life⁽¹²⁹⁾. Most of spectrophotometric methods developed for the determination of cobalt

are, unfortunately, not practically usable due to poor sensitivity and partly poor selectivity. Routine spectrophotometric methods are not often sensitive enough to determine low concentrations of cobalt ions in environmental samples only at the $\mu\text{g/l}$ level. Consequently, a pre concentration step is usually required. In spite of recent advances in instrumental analysis, a direct determination of trace elements in complex matrices, such as environmental, biological, mineral, ore and high purity materials, seems still to be difficult because of insufficient sensitivity and selectivity of the methods. Thus, enrichment and separation of the analytes are important for the determination of Co(II), because of its extremely low concentration in natural water. Cobalt determination at trace levels in real samples has been frequently difficult because of low concentration of the metal and matrix interferences⁽¹³⁰⁾. Flame atomic absorption spectrometry (FAAS) is one of the techniques most extensively used for determining various elements with a significant precision and accuracy. This analytical technique is remarkable for its selectivity, speed and fairly low operation cost. However, in some cases it is rather difficult to determine traces of heavy metals in environmental samples due to insufficient sensitivity or matrix interferences. Thus, a preconcentration and/or separation step is necessary⁽¹³¹⁾. Nowadays, Cloud Point Extraction (CPE) using non-ionic surfactants has attracted considerable attention as an alternative to the conventional extraction techniques for separation and preconcentration⁽¹³²⁾. It is well known that CPE used surfactant has the potential to intensify solubility of hydrophobic materials, extraction technology which is environment complaisant to many researchers and luring many analytical chemists as to other extraction system. Table

(1.4): compresses the uses of CPE for assurance of Co(II) in various matrices.

Table (1.4): Cloud point extraction of Cobalt in different matrixes

Sample type	Chelating agent	Surfactant type	Detection system	PF	LOD	Ref.
Water and B12,B-complex vitamins	MPMP	Triton X-114	FI-FAAS	-	0.21 $\mu\text{g. L}^{-1}$	128
Water samples	BTANP	Triton X-114	FAAS	50	1.5 ng.ml^{-1}	133
Water samples	MOSDAA	Triton X-114	FAAS	19	0.47 ng.ml^{-1}	134
Tap, sea ,fish , black tea	1-PTSC	Triton X-114	FAAS	25	1 $\mu\text{g.L}^{-1}$	135

1.11: Factors Affecting Cloud Point Extraction

The performance of a CPE process is influenced by many factors, such as surfactant type and its concentration, ionic strength and equilibration temperature and time .

1.11.1: Type and concentration of surfactant

Surfactant concentration plays an important role in phase separation as lower concentration makes the separation to be incomplete while higher concentration than the desired amount enhances the surfactant-rich phase's viscosity which concurrently decreases its volume. Increasing the concentration of the surfactant also increases a number of

Surfactant aggregate complexes⁽¹³⁶⁾. Explicitly, surfactant plays a vital role in the CPE process. Thus, proper selection of surfactant could lead to satisfactory performance of the CPE process. There are several types of surfactants used in the CPE processes include Triton series and PONPE series (polyethyleneglycol mono-4-nonylphenyl ethers). However, these surfactants contain alkyl phenyl groups in their hydrophobic moiety, leading to some environmental concerns. To overcome this problem, biodegradable surfactant, mainly polyethylene glycol or silicone surfactant without phenyl group is proposed⁽¹³⁷⁾. The main requirements for a surfactant to be selected as being suitable for the CPE experiments are associated mainly with its physicochemical characteristics: high density, which facilitates phase separation, low cloud point temperature, below or near room temperature in a narrow concentration range, for thermo labile molecules, balanced hydrophobicity, commercial availability and low cost⁽¹³⁸⁾. Depending on the nature of the hydrophilic group, surfactants are classified as non-ionic, zwitterionic, cationic, and anionic. Nowadays, non-ionic, zwitterionic and anionic surfactants are most widely used in the CPE process for organic compounds, drugs, biomaterials and inorganic metal ions. However, the application of the cationic surfactants in the CPE is scarce. Since, it can directly affect the extraction and preconcentration, and accuracy of the final analytical results, it is very important to select an appropriate surfactant for a successful CPE analysis⁽¹³⁹⁾.

1.11.2: Effect of ionic strength

Generally, clouding is a phenomenon detected in non-ionic surfactants when the solution is heated to a threshold temperature which is known as clouding point temperature (CPT). The clouding phenomenon is uncommon with the ionic surfactants, presumably because of the large electrostatic repulsion between the aggregates that prevents phase separation in most cases. The phase separation occurs within a narrow temperature range into surfactant rich phase and aqueous phase, because of density difference due to a sharp increase in aggregation number of the micelles and the decrease in intermicellar repulsion. The CPT has been found extremely dependent on the presence of additives⁽⁸⁷⁾. Usually, the CPT of a non-ionic surfactant can be altered in the addition of additives, such as inorganic salt and organic compounds, into an aqueous solution. This phenomena is known as salting-out effect where the addition of neutral salts depresses the CPT of a nonionic surfactant aqueous solution in proportion to their concentration⁽¹⁴⁰⁾. Several factors have been considered to be responsible for the CPT phenomenon additives in the such as surfactant molecule and its system of CPE. However, CPT is very sensitive to the presence of additives used in the CPE system, even at a very low concentration. The additives modify the surfactant-solvent interactions, change the CMC, size of micelles and phase behavior in the surfactant solutions. Many efforts has been made to investigate the effect of various additives e.g. inorganic electrolytes, organic compounds, ionic surfactant, cationic surfactant and zwitterionic surfactants on the cloud point of a nonionic surfactant⁽¹⁴¹⁾.

1.11.3: Equilibration temperature and time

The optimum incubation time and equilibrium temperature are necessary to achieve an easy and efficient phase separation with preconcentration. The buildup of surfactant-rich phase are solely subjected by demeanor of the employed surfactant⁽¹⁴²⁾. Theoretically, the phase separation can be achieved once the optimal equilibration temperature of the CPE which is higher than the CPT of the surfactant. If the temperature is lower than the CPT, two phases cannot be formed.

However, too high temperature may lead to the decomposition of analytes. It has also been demonstrated that the analyte extraction efficiency and preconcentration in the CPE increase as the equilibration temperature for phase separation is progressively increased to above the CPT. Similarly, as the equilibration temperature increases; the volume of surfactant rich phase decrease because the hydrogen bonds are disrupted and dehydration occurs. Based on a physical point of view, the increase of temperature provokes the dehydration of the hydrophilic groups of the surfactant molecules, an increase of the aggregation number and the swelling of the micelles until the micellar solution become turbid and the separation of the surfactant rich phase takes place⁽¹⁴³⁾. Since longer equilibration times (> 30 min) do not have any significant effect on the extraction, the equilibration time of 10 – 20 min is sufficient to obtain good extraction in most work⁽¹³⁹⁾.

1.12: Application of Cloud Point Extraction

The verification of the heavy metals in environmental samples is of vital importance which has received much attention by researchers in the last year, in order to pinpoint their impact. the applications of CPE for the selected metals .

1.12.1: Water

Water is important for the existence, development and preservation of all human life, making it an important commodity in the world. However, nearly one billion people in the world lack access to safe drinking water, mainly those living in third world countries⁽¹⁴⁴⁾. Water is an essential to maintain good health and proper body functions. Doctors recommend drinking of 1.9 to 2.1 liters of water daily. This quantity is necessary because water occupies approximately 60% of a person's body weight and it is the most abundant compound in human body. This provides a suitable environment for vital bio-chemical reactions in the body as water is the dispersing media⁽¹⁴⁵⁾. Heavy metals in the environment are really increasing due to human activity, particularly mining process and heavy industry in the developing world. This has caused great accumulation of high concentrations of toxic heavy metals in natural waters⁽¹⁴⁶⁾. Drinking water is also an important source for heavy metals for humans. The levels of heavy metal ions in drinking water are generally at $\mu\text{g/l}$. For precise and accurate analysis of heavy metal ions in drinking water, it is possible to use inductively coupled plasma-mass spectrometry (ICP-MS), inductively coupled plasma-atomic emission spectrometry (ICP-AES) and graphite furnace atomic absorption spectrometry (GFAAS). However, these techniques are expensive and difficult to use. The separation/preconcentration- flame atomic absorption spectrometry (FAAS) combination is an important alternative for these techniques due to cheap cost and easy usage of FAAS. Concern has increased over the concentration of trace metal ions in drinking water.

Organizations like the World Health Organization and the European Committee recommend limits for drinking water that require much greater sensitivity in measurement than is obtainable by flame atomic absorption spectrometry. Preconcentration- separation techniques like coprecipitation, liquid-liquid extraction, cloud point extraction and ion exchange are used prior to flame atomic absorption spectrometric determination of heavy metals in drinking waters⁽¹⁴⁷⁾.

1.12.2: Milk

Milk is considered as nearly a complete food in that it's a good source of protein, fat, and major minerals⁽¹⁴⁸⁾. Cow milk contains some major elements such as calcium, potassium, phosphorous, and magnesium in addition to sodium, chlorine and even heavy metals. Increase in industrial and agricultural processes have resulted in increased concentration of metals in the air, water and soil. These metals are taken in by plants and consequently accumulate in their tissues. Animals that graze on such contaminated plants and drink from polluted waters also accumulate such metals in their tissues and milk if lactating⁽¹⁴⁹⁾. A large amount of these metals taken in by plants and animals subsequently find their way into the food chain. This ever increasing pollution has given rise to concern on the intake of harmful metals in humans. Metals enter the human body through inhalation, ingestion or absorption through the skin⁽¹⁵⁰⁾. In recent times, the amount of metals in cow milk is widely studied, particularly in industrialized and polluted areas of the developed and the developing countries of the world⁽¹⁵¹⁾. Although some metals are essential nutrients, which have a variety of biochemical functions in all

living organisms and important industrial uses, their potential toxicity to humans and animals is a source of concern. It is, therefore, necessary to monitor and control their levels in consumed food. The measurement of metal levels is helpful not only in ascertaining risk to human health but also in the assessment of environmental quality⁽¹⁵²⁾. Many reports indicated heavy metals in milk and attributed the presence of these heavy metals in milk to exposure of cows to environmental pollution, consumption of contaminated feed stuffs and water as well as the production process⁽¹⁵³⁾. Milk is the essential food needed by growing children. It contains both the basic and additional requirements needed by children especially during their developmental years. This milk may be contaminated by heavy metals that at certain levels may cause toxicity to consumers. This toxicity is attributed to accumulation of heavy metals in the body. The most commonly method for metal analysis is flame Atomic absorption spectrometry. Here, the interference by other elements is almost nil or negligible because the estimation is carried out for an element at a specific emission spectra line. In fact, in Atomic absorption spectrometry, traces of one element can be determined accurately in the presence of a high concentration of other element⁽¹⁵⁴⁾.

1.12.3: Tobacco

Tobacco plant is amenable to absorb and accumulate heavy metal species from the soil into its leaves⁽¹⁵⁵⁾, or deposited on tobacco leaves from air⁽¹⁵⁶⁾. Cigarette is made up of tobacco, paper and additives. As much as 600–1400 additives are used in cigarette manufacture, with many of these additives containing environmental contamination and exposure to heavy metals such as mercury, cadmium and lead is a serious

growing problem throughout the world⁽¹⁵⁷⁾. It is known that the content of metals in tobacco varies a lot and it depends on several factors such as soil type and pH, use of a metal-containing pesticides, fertilizers, etc. Some of the metals such as iron, manganese, zinc and copper are very important for plant growth. However, other metals such as lead, cadmium and nickel are not important for the plant growth, but they can cause serious health and ecological problems. In both cases, we need to pay attention to their right determination. It is known that the latter depends on the right selection of a method for test sampling and suitable measuring equipment. Although, a number of investigations have been carried out on heavy metal concentration in tobacco leaves⁽¹⁵⁸⁾, There are several studies reported in the literature on heavy metal contents in the cigarettes sold and smoked in different parts of the world. A. Pourkhabbaz, and H.Pourkhabbaz⁽¹⁵⁹⁾ have investigated the levels of toxic metals (Cd, Cu, Co Ni, Zn and Pb) in the tobacco of different Iranian cigarette brands and related health issues. Ashraf⁽¹⁶⁰⁾ has reported levels of Cd and Pb in popular cigarette brands sold and/or produced in Saudi Arabia. Level of heavy metals (Cr, Cd and Pb) in selected cigarettes and tobacco leaves in Benue State. Thus, cigarette tobacco is very harmful, toxic and genotoxic for human health⁽¹⁶¹⁾. Therefore, the concentration of heavy metals in cigarette brands is of importance because of its toxicological effects⁽¹⁵⁷⁾.

1.12.4 : Tea

Tea is one of the most popular nonalcoholic beverages, consumed by over two-thirds of the world's population for its medicinal, refreshment and mild stimulant effects⁽¹⁶²⁾. Tea leaves contain polyphenols which has many medicinal properties, including antioxidant⁽¹⁶³⁾, cholesterol lowering⁽¹⁶⁴⁾ and anticancer activities. However, considering that an estimated 18 billion cups of tea are consumed daily worldwide⁽¹⁶⁵⁾. Therefore, tea should be free from contaminants such as heavy metals, which are toxic and harmful to the human body because of their non-biodegradable nature and persistent accumulation in different body parts⁽¹⁶⁶⁾. Prolonged consumption of heavy metals from food can lead to their accumulation in the kidney and liver, causing disruption of numerous biochemical processes and potentially causing cardiovascular, nervous and bone diseases⁽¹⁶⁷⁾. Elemental analysis of a tea sample requires destruction of the organic fraction of the sample, leaving the heavy metals either in solution or in a form that is readily dissolved. Unfortunately, because of a large number of analytes and a variety of sample types, there is no universal sample preparation technique that meets all of the diverse requirements. Among the strategies for sample preparation, dilution, acid digestion and extraction are the most commonly considered^(168,169).

1.12.5: Soil

High concentrations of heavy metals may affect the ecosystem and human health. Heavy metals may be transported through the food chain to the human body and have a significant toxicity to people. The environmental issues related to heavy-metal contamination are becoming

serious in developing countries⁽¹⁷⁰⁾. With the rapid industrialization and urbanization trend, the increment of traffic activities substantially contributes to the accumulations of heavy metals discharged by vehicles in roadside environments. Heavy-metal pollution in agricultural areas owing to traffic emissions may contaminate the crops growing near the roadways⁽¹⁷¹⁾. In agricultural areas, uptake of heavy metals through the soil-crop system could play a role in human exposure to heavy metals⁽¹⁷²⁾. Cu, Zn, Cd, and Pb are the typical metal pollutants due to traffic activities⁽¹⁷³⁾. At low doses some heavy metals are essential micronutrients for plants, but in higher doses, they may cause metabolic disorders and growth inhabitation in most plant species⁽¹⁷⁴⁾. On the other hand, Cd even at extremely low concentrations, is toxic and lead to many diseases, including increased risk of cancer⁽¹⁷⁵⁾. Cd emission is mainly from lubricating oil consumption and tire wear. Brake wear is the most important source for Cu emissions. Through the atmospheric deposit or road runoff, heavy metals can be transported into the roadside soils⁽¹⁷⁶⁾. On average, the heavy metals' concentrations in roadside grasses are significantly lower than those in roadside soils^(177,178). Observation studies have been conducted in many countries to investigate the heavy-metal contamination of roadside soils. It was found that the heavy-metal concentrations were influenced by multiple factors, such as traffic volume⁽¹⁷⁹⁾, highway characteristics⁽¹⁸⁰⁾, road and roadside terrain⁽¹⁸¹⁾, rainfall⁽¹⁷⁹⁾, High traffic volume can heighten the heavy-metal content in the roadside .

1.13: Aim of the study

1- Synthesis of azo-azomethine dyes and their using of cloud point extraction for metal ions .

2- Characterization of azo-azomethine dyes in several ways, including UV-Visible, FT- IR, ^1H -NMR and Mass spectroscopy.

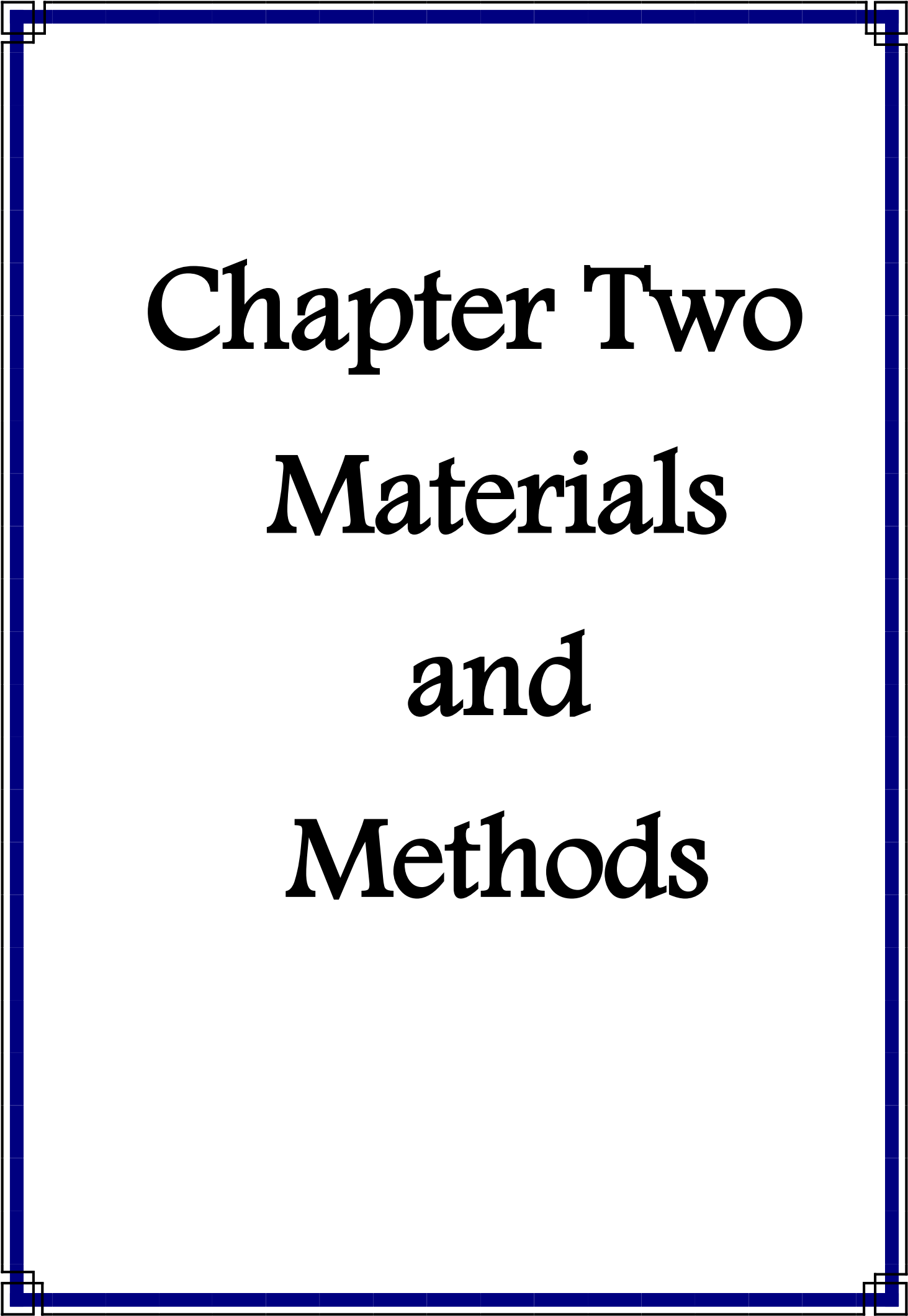
3- Study primary tests for reaction between metal ions and reagents and determination optimization of the experimental conditions for formation complexes. Also, preparation and characterization of azo-azomethine complexes .

4- Most of the works, done by CPE in metals analysis, deal with the determination of an analyte as metal ions in different matrices. Hence, there are an obvious lacking for using CPE in the extraction and enrichment of metal ions as metal anion complexes or species (i.e. inorganic anions) which would possibly be found in the environmental samples in these forms. Also, there are insufficient reports to judge on the eligibility of CPE in the separation and preconcentration of dyes in the complex matrices. Therefore, the main goal of this study is to open new routes and attempts in order to broaden the analytical characteristics of CPE methodology by FAAS and its applicability in different fields.

This was through the following objectives;

- a) Finding the optimum conditions for the separation and enrichment of the metal ions (Cu^{2+} , Cd^{2+} and Co^{2+}), that is, concentration of reagent, HCl concentration, incubation time, equilibration temperature and Triton X-100 amount were selected in this study, in an attempt to improve the selectivity and the precision of the analytical results.

- b) Studying the separation mechanisms for the selected species thermodynamically and kinetically through the finding of the equilibrium constants for the extraction and thermodynamic parameters such as ΔH , ΔG and ΔS .
- c) Studying the accuracy, sensitivity and efficiency of the method by calculation EF, PF, LOD, LOQ, %RSD and CI.
- d) To apply the developed method in this thesis to detect the pollutants by determination concentration of metal ion in water (river and tap), milk, tea, tobacco and soil .



Chapter Two

Materials

and

Methods

Materials and Methods

2.1: Instrumentation

The main instruments used in this research project are as follows:

2.1.1: UV-Vis Spectrophotometric Measurements

A Shimadzu double beam UV-Vis Spectrophotometer model UV-1700 (Japan) was used for the scanning of absorption spectra of all reagents and complexes throughout this study. While absorbance measurements in the optimization study and detection of metals were done with Single beam (UV-Vis) Spectrophotometer, T60, PG-instruments Ltd has been measuring the spectra of solutions at room temperature using quartz cells width (1cm) in the laboratories of the department of chemistry, college of science, university of Thi - Qar.

2.1.2: Fourier Transform Infrared Spectrophotometer

The FT-Infrared measurements for all reagents and metal complexes prepared in this study were recorded by using FT-IR affinity Spectrophotometer (Shimadzu) Japan in the department of chemistry, college of science, university of Thi - Qar.

2.1.3: Flame Atomic Absorption Spectrophotometric Measurements

A double-beam Atomic Absorption Spectrophotometer AA-500AFG (Shimadzu, Japan) is equipped with burner unit made of titanium (10 cm slot) air-cooled premixed for air/acetylene flame.

2.1.4: ^1H -NMR spectra

^1H -NMR spectra of compounds are recorded in Tehran University, Iran. The chemical shift values are expressed δ (ppm), using tetramethylsilane (TMS) as internal standard .

2.1.5: Mass spectra

Mass spectra are recorded of compounds using Agilent Technology (HP)/ MS (EI,70ev), Model 5973 Network Mass Selective Detector in the university of Tehran , Iran.

2.1.6: Molar Electrical Conductivity

Determine molar conductivity for all the complexes prepared in dimethylsulfoxide (DMSO) solvent using inolabcond devise 720 and pole platinum was fixed cell user is equal to 1.52cm^{-1} and a concentration of 10^{-3} M at room temperature.

2.1.7: Other Supplementary Equipments Used in This Work Were:

A- pH- meter: WTW, E163694, (Germany).

B- Electrical balance; Germany – Artorius,1320

C- Electrical furnace: Japan – Hirayama 2004

D- Electrical furnace: Oven Philip Harris Limited, Shenstone, Ser. No. T 7981S1 (England).

E- TLC: Thin layer chromatography is performed using TLC grade silica gel .

F- Water bath: Thomas Sc. Ap. (England) was used to control the desired temperature so as to achieve the phase separation .

G-Melting point Measurement: The melting grades of the prepared compounds were measured using a type of device point/SMP31 Melting .

2.2 : Chemicals

All chemicals used in this study were presented in table (2.1) and used directly without any further purification .

Table (2.1): Chemicals used in this work

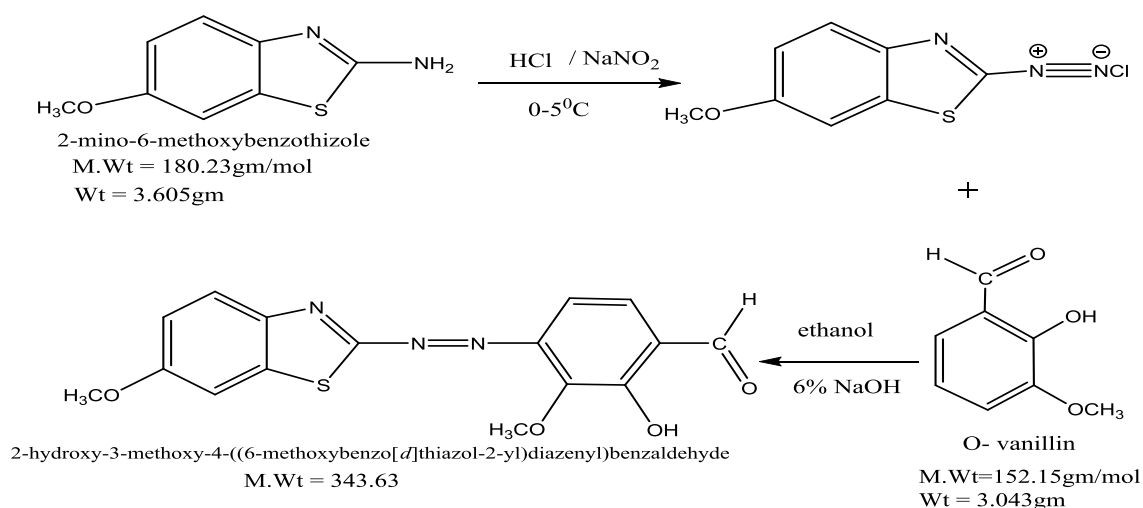
Substance	Company Name	Chemical Formula	Molecular weight (g/mol)	Purity %
Acetic acid (glacial)	BDH	C ₂ H ₄ O ₂	60.05	99.5
3,4-diaminotoluene	BDH	C ₇ H ₁₀ N ₂	122.17	97.0
2-amino-6-methoxybenzothiazole	BDH	C ₈ H ₈ N ₂ SO	180.23	99.0
Aniline	BDH	C ₆ H ₇ N	93.13	99.0
O-toluidine	BDH	C ₇ H ₉ N	107.15	
2-aminobenzylamine	Sigma Aldrich	C ₇ H ₁₀ N ₂	122.17	98.0
O-phenylenediamine	Sigma Aldrich	C ₆ H ₈ N ₂	108.14	99.5
O-Vanillin	Sigma Aldrich	C ₈ H ₈ O ₃	152.15	99.0
Salicylaldehyde	Sigma Aldrich	C ₇ H ₆ O ₂	122.12	99.0
Copper (II) chloride dihydrate	BDH	CuCl ₂ .2H ₂ O	170.48	99.0
Cadmium chloride hydrate	BDH	CdCl ₂ .H ₂ O	201.32	99.0
Cobalt (II) Chloride Hexahydrate	BDH	CoCl ₂ .6H ₂ O	237.93	97.0
Nickel (II) Chloride Hexahydrate	BDH	NiCl ₂ .6H ₂ O	237.69	98.0
Chromium (III) Chloride Hexahydrate	BDH	CrCl ₃ .6H ₂ O	266.45	99.0
Methanol	BDH	CH ₃ OH	32	99.0
Ethanol	BDH	C ₂ H ₅ OH	46.07	98.0
Hydrochloric acid	BDH	HCl	36.5	37.0
Hydrogen peroxide	Fluka	H ₂ O ₂	34.0147	50.0
Per chloric acid	BDH	HClO ₄	100.47	75.0
DMSO	G.C.C	C ₂ H ₆ O _S	78.13	99.5
Nitric acid	BDH	HNO ₃	63.01	69.0
Sodium hydroxide	Fluka	NaOH	40	99.0
Sodium nitrite	BDH	NaNO ₂	68.9953	99.0
TritonX-100	Sigma Alidrich	C ₈ H ₁₇ C ₆ H ₄ (OC ₂ H ₄) _n	625	99.6

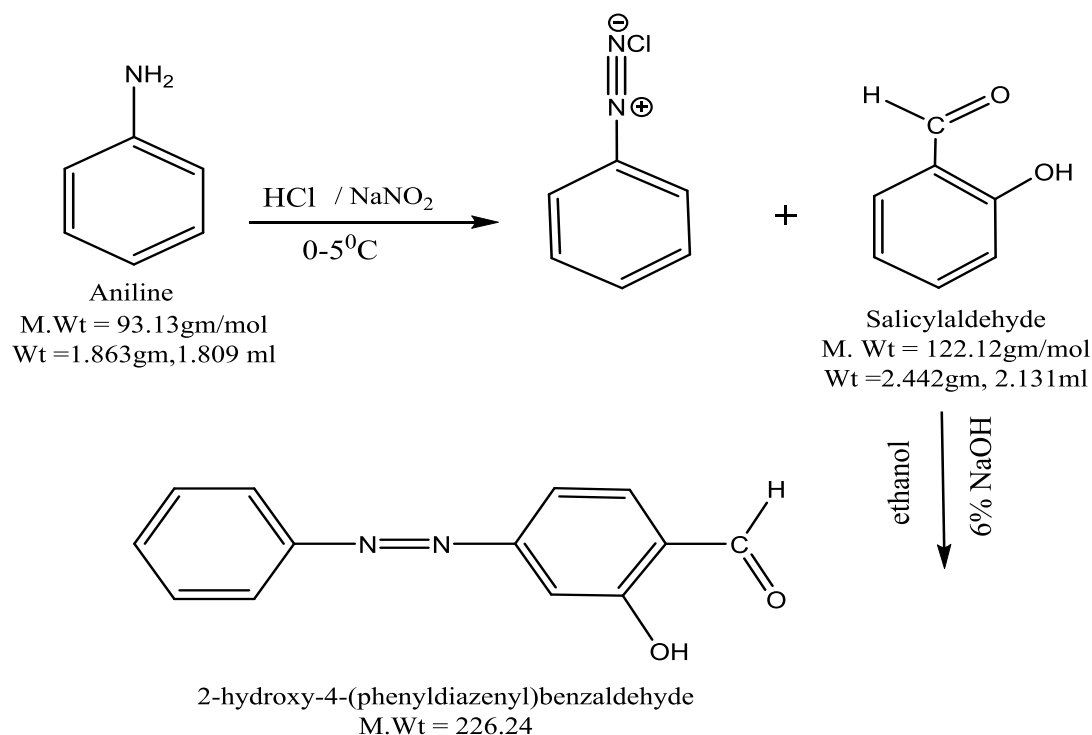
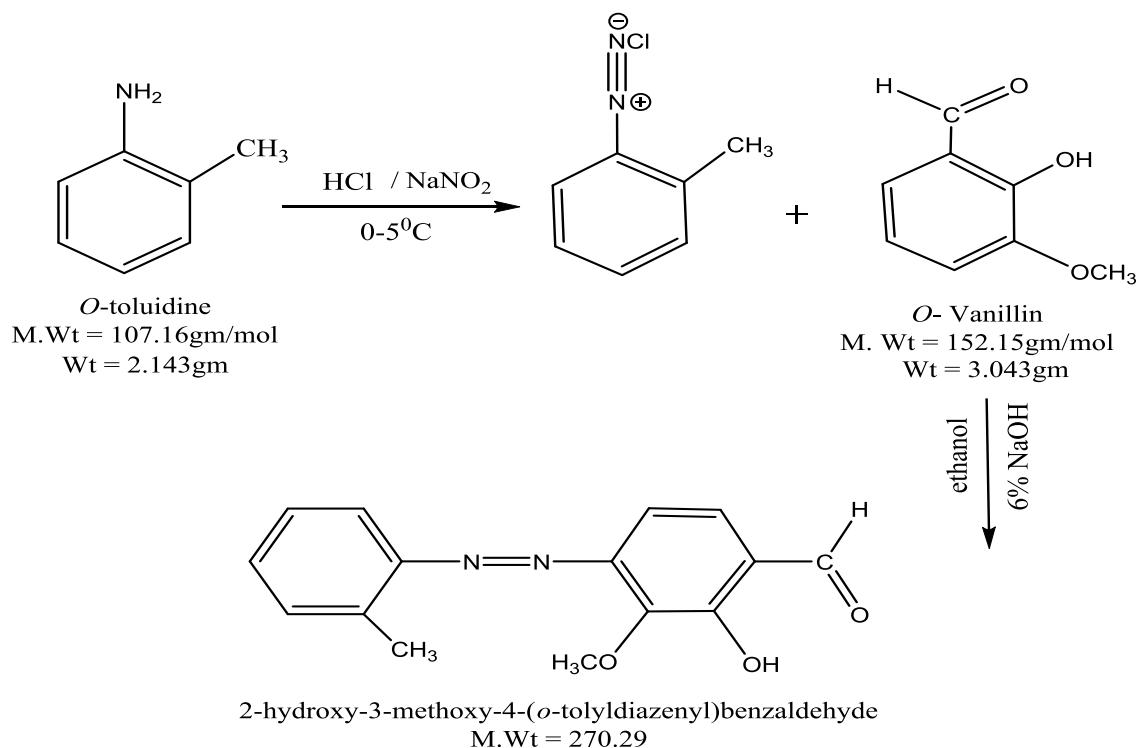
2.3 : Synthesis Methods

2.3.1: General procedure of azo dyes

In general, the Azo dyes prepared by the reaction of the (0.02mole) amine was dissolved in mixture 30ml distilled water and 5ml of concentrated hydrochloric acid and cooled to (0-5) °C. The solution was diazotized at (0-5) °C with (1.38gm, 0.02mole) sodium nitrate was dissolved in 20ml distilled water was added drop wise to amine solution and stirring continuously at (0-5)°C and left to stand 30 min .This diazonium solution was added to aldehyde (0.02mole) was dissolved in 25ml ethanol and 30 ml of 6% sodium hydroxide. The mixture was stirred continuously for 2h. at (0-5) °C in ice-bath, acidified with dilute hydrochloric acid to pH = 6 and allowed to stand in the refrigerator overnight. The precipitate was filtered and washed with distilled water and ethanol solution to remove the excess of unreacted substances and recrystallized from ethanol and dried in oven at 50 °C for two hours⁽¹⁸²⁾. The purity was confirmed by TLC technique.

2.3.1.1: 2-hydroxy-3-methoxy-4-((6-methoxybenzo[d]thiazol-2-yl) diazenyl)benzaldehyde (Az₁)



2.3.1.2: 2-hydroxy-4-(phenyldiazenyl)benzaldehyde(AZ₂)2.3.1.3: 2-hydroxy-3-methoxy-4-(o-tolyldiazenyl)benzaldehyde(AZ₃)

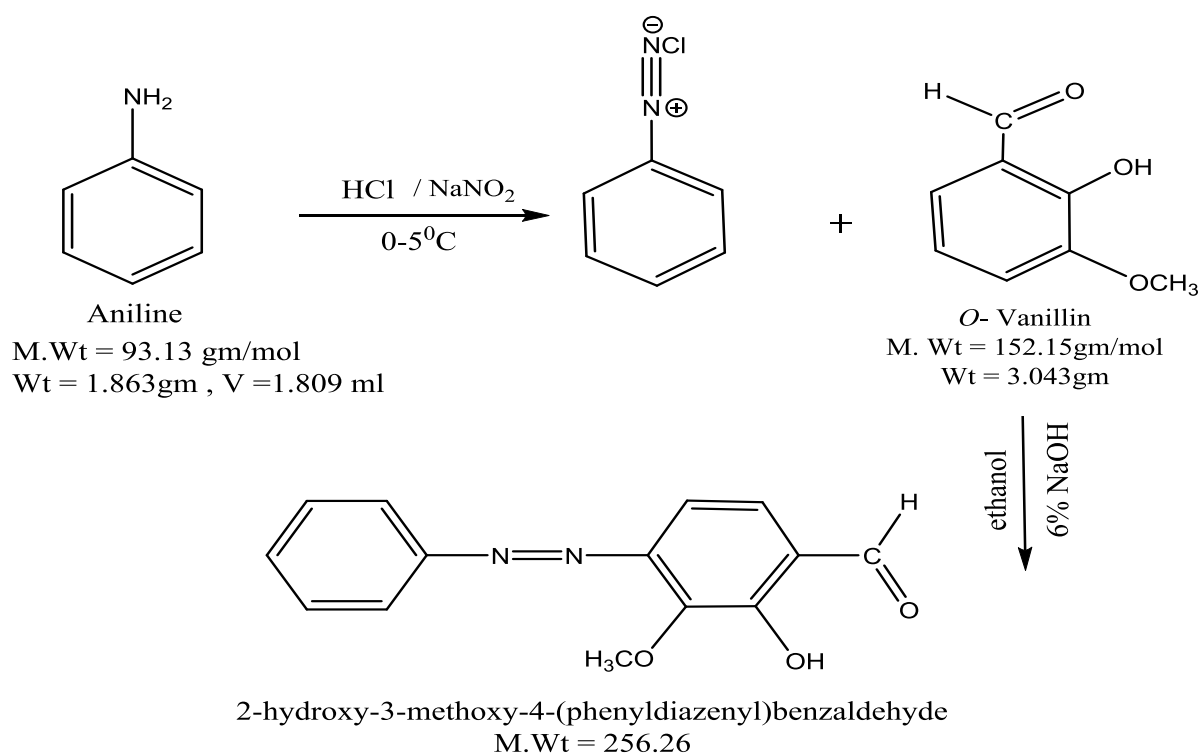
2.3.1.4: 2-hydroxy-3-methoxy-4-(phenyldiazenyl)benzaldehyde(AZ₄)

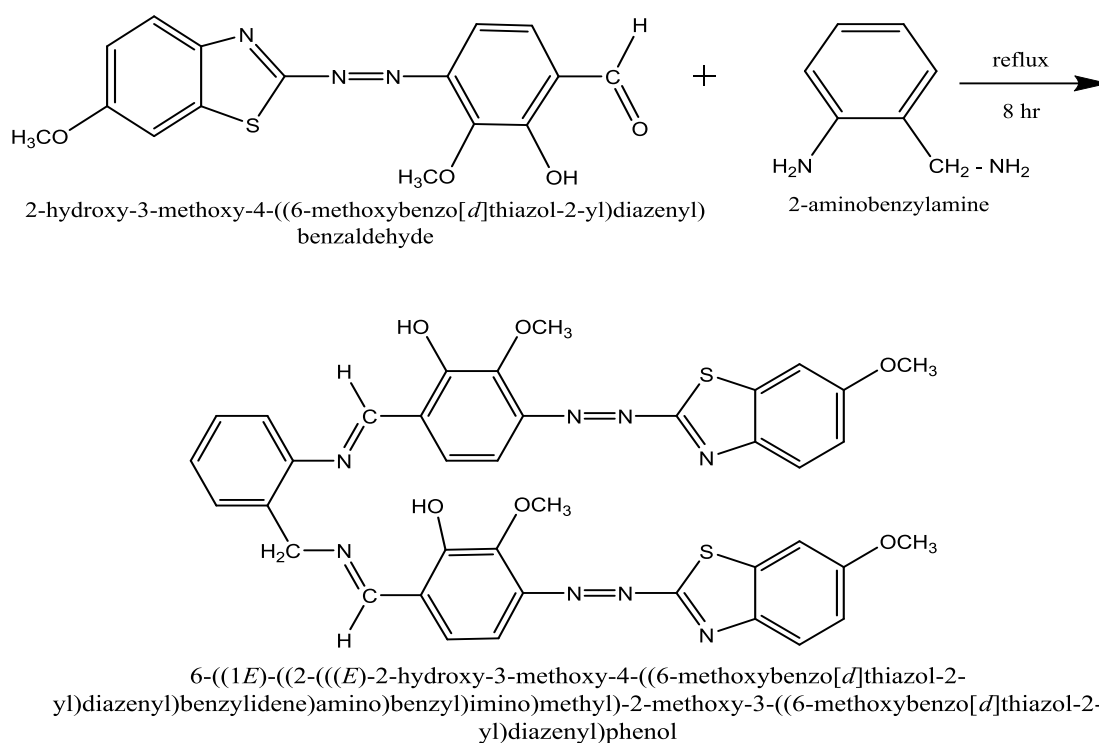
Table (2.2): Some physical and chemical properties of azo dyes

Comp.	Chemical formula	M.Wt	Color	m.p°C	Yield%
AZ ₁	C ₁₆ H ₁₃ N ₃ O ₄ S	343.63	Reddish orange	179-180	82.64
AZ ₂	C ₁₃ H ₁₀ N ₂ O ₂	226.24	Yellow brown	159-160	85.06
AZ ₃	C ₁₅ H ₁₄ N ₂ O ₃	270.29	Orange	142-144	80.37
AZ ₄	C ₁₄ H ₁₂ N ₂ O ₃	256.26	Light orange	133-135	79.50

2.3.2: Synthesis of Azo-Azomethine Ligands

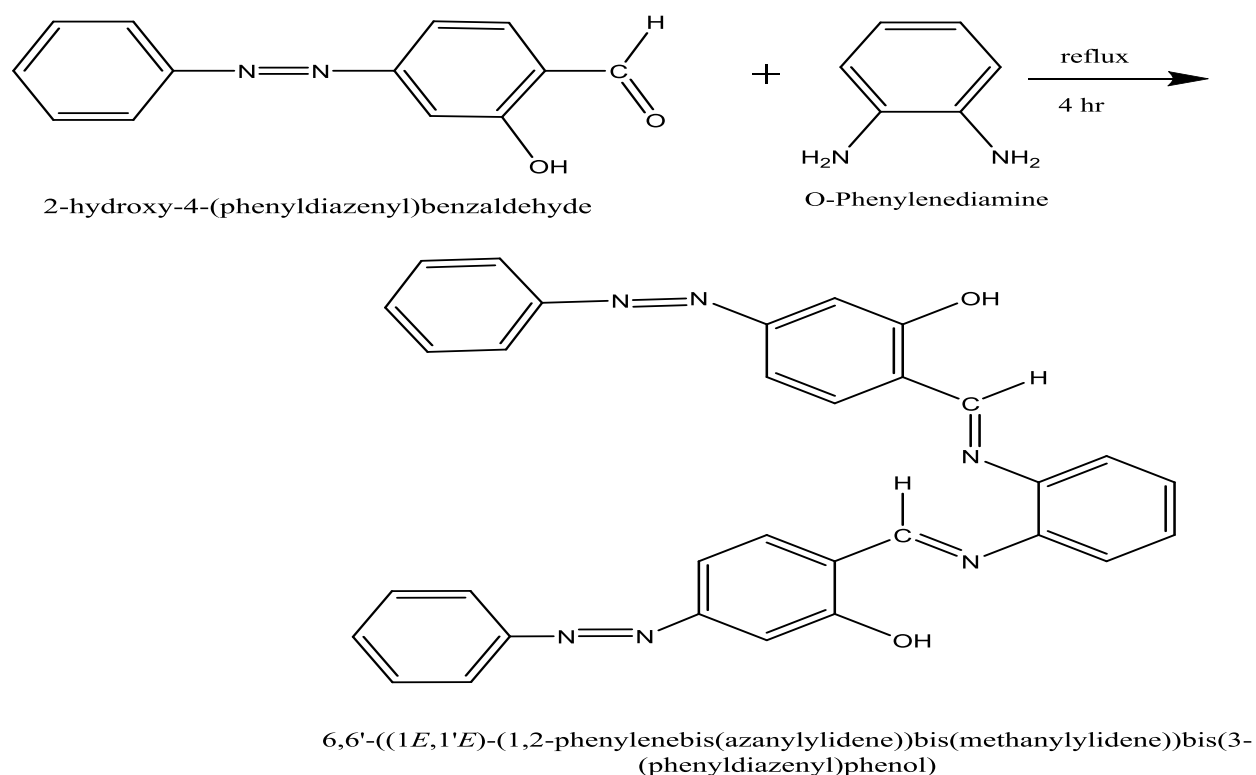
2.3.2.1: Synthesis of 6-((1E)-((2-(((E)-2-hydroxy-3-methoxy-4-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)benzylidene)amino)benzyl) imino)methyl)-2-methoxy-3-((6-methoxybenzo[d]thiazol-2-yl)diazenyl) phenol (L_1)

The azo-azomethine ligand was synthesized by condensation reaction of 2-aminobenzylamine with azo dye compound **2-hydroxy-3-methoxy-4-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)benzaldehyde (AZ_1)**, by dissolving (3.436gm, 0.01mole) of (AZ_1) in (20ml) absolute methanol then mixed with a solution (0.611gm, 0.005mole) of (2-aminobenzylamine) dissolved in (10ml) of the same solvent with the addition of four drops of glacial acetic acid as a catalyst followed by reflux for (8) hours. The reaction was followed using TLC⁽¹⁸³⁾. The light brown precipitate was formed, It was filtered ,dried and recrystallized in absolute ethanol .



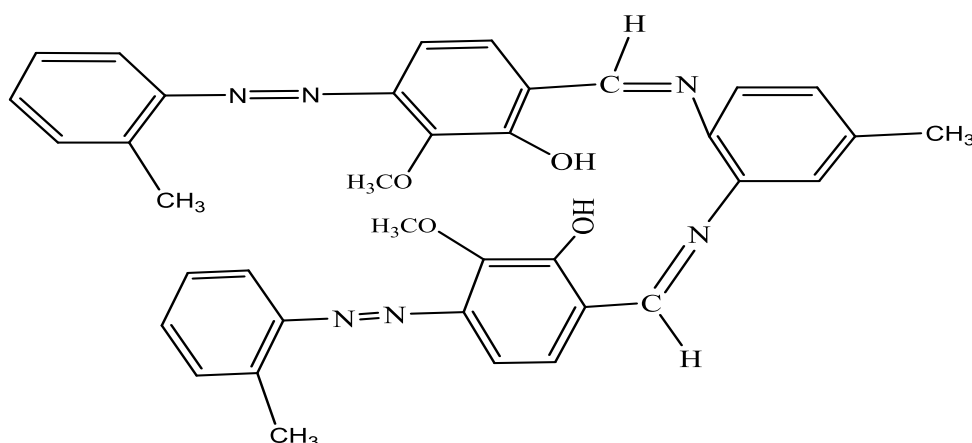
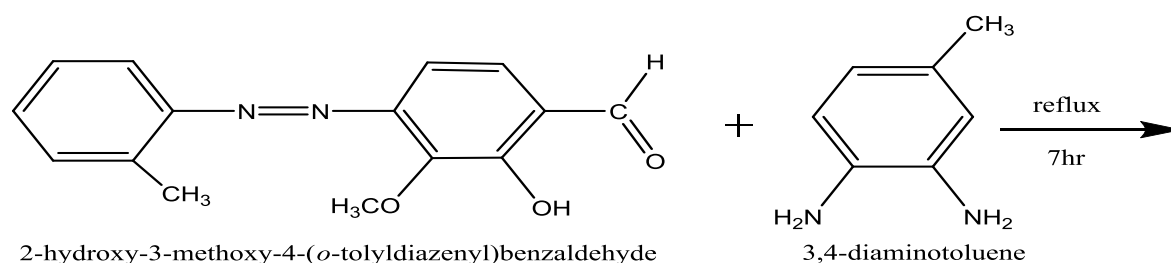
2.3.2.2:6,6'-((1E,1'E)-(1,2-phenylenebis(azanylylidene))bis(methanylylidene))bis(3-(phenyldiazenyl)phenol)(L₂)

The azo-azomethine ligand was synthesized by condensation reaction of O-phenylenediamine with azo dye compound **2-hydroxy-4-(phenyldiazenyl)benzaldehyde (AZ₂)**, by dissolving (2.262gm, 0.01 mol) of (AZ₂) in (20ml) absolute methanol then mixed with a solution (0.541gm, 0.005mol) of (O- phenylenediamine) dissolved in (10ml) of the same solvent with the addition of four drops of glacial acetic acid as a catalyst followed by reflux for (4) hours. The reaction was followed using TLC⁽¹⁸³⁾. The brown precipitate was formed, It was filtered, dried and recrystallized in absolute ethanol .



2.3.2.3:6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene))bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol) (L₃)

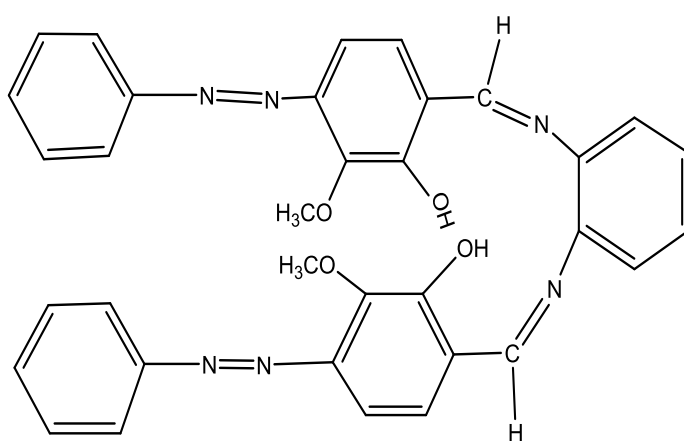
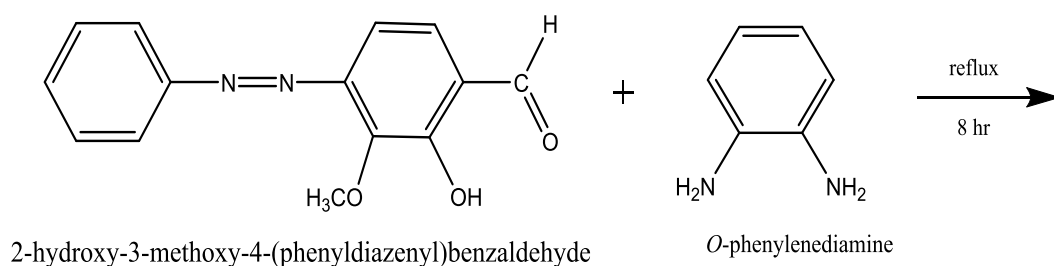
The azo-azomethine ligand was synthesized by condensation reaction of 3,4-diaminotoluene with azo dye compound **2-hydroxy-3-methoxy-4-(o-tolyldiazenyl)benzaldehyde (AZ₃)**, by dissolving (2.703gm, 0.01mol) of (AZ₃) in (20ml) absolute methanol then mixed with a solution (0.611gm, 0.005mol) of (3,4-diaminotoluene) dissolved in (10ml) of the same solvent with the addition of four drops of glacial acetic acid as a catalyst followed by reflux for (7) hours . The reaction was followed using TLC⁽¹⁸³⁾. The dark brown precipitate was formed, It was filtered ,dried and recrystallized in absolute ethanol .



6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene))bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol)

2.3.2.4:6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene)) bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol) (L₄)

The azo-azomethine ligand was synthesized by condensation reaction of O-phenylenediamine with azo dye compound **2-hydroxy-3-methoxy-4-(phenyldiazenyl)benzaldehyde (AZ₄)**, by dissolving (2.563gm, 0.01mol) of (AZ₄) in (20ml) absolute methanol then mixed with a solution (0.541gm, 0.005mol) of (O-phenylenediamine) dissolved in (10ml) of the same solvent with the addition of four drops of glacial acetic acid as a catalyst followed by reflux for (8) hours. The reaction was followed using TLC⁽¹⁸³⁾. The brown precipitate was formed, It was filtered ,dried and recrystallized in absolute ethanol .



6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene)) bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol)

Table (2.3): Some physical and chemical properties of the azo-azomethine compounds

Comp.	Chemical formula	M.Wt	Color	m.p°C	Yield%
L ₁	C ₃₉ H ₃₂ N ₈ O ₆ S ₂	772	Light brown	288-290	80.37
L ₂	C ₃₂ H ₂₄ N ₆ O ₂	524	Brown	210-212	80.00
L ₃	C ₃₇ H ₃₄ N ₆ O ₄	626	Reddish orange	253-254	78.13
L ₄	C ₃₄ H ₂₈ N ₆ O ₄	584	Dark brown	194-196	79.57

2.4: Preparation of Standard Solutions of metal ions and Reagents

2.4.1: Stock Solution of Cd(II) as CdCl₂.H₂O (1000 µg mL⁻¹)

This solution was prepared by accurately weighing 0.1791gm of CdCl₂.H₂O into a 50 ml beaker, added minimum amount of deionized water and swirled to dissolve. The content was transferred quantitatively to a 100 ml volumetric flask, and then diluted to the mark with deionized water .

2.4.2: Stock Solution of Cu(II) as CuCl₂.2H₂O (1000 µg mL⁻¹)

This solution was prepared by accurately weighing 0.2682gm of CuCl₂.2H₂O into a 50 ml beaker, added minimum amount of deionized water and swirled to dissolve. The content was transferred quantitatively to a 100 ml volumetric flask, and then diluted to the mark with deionized water .

2.4.3: Stock Solution of Co(II) as $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ ($1000 \mu\text{g mL}^{-1}$)

This solution was prepared by accurately weighing 0.4038gm of $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ into a 50 ml beaker, added minimum amount of deionized water and swirled to dissolve. The content was transferred quantitatively to a 100 ml volumetric flask, and then diluted to the mark with deionized water .

2.4.4: Stock Solution of Ni(II) as $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$ ($1000 \mu\text{g mL}^{-1}$)

This solution was prepared by accurately weighing 0.4049gm of $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ into a 50 ml beaker, added minimum amount of deionized water and swirled to dissolve. The content was transferred quantitatively to a 100 ml volumetric flask, and then diluted to the mark with deionized water .

2.4.5: Stock Solution of Cr(III) as $\text{CrCl}_3 \cdot 6\text{H}_2\text{O}$ ($1000 \mu\text{g mL}^{-1}$)

This solution was prepared by accurately weighing 0.5124gm of $\text{CrCl}_3 \cdot 6\text{H}_2\text{O}$ into a 50 ml beaker, added minimum amount of deionized water and swirled to dissolve. The content was transferred quantitatively to a 100 ml volumetric flask, and then diluted to the mark with deionized water .

2.4.6: Metal ions standard solutions ($100 \mu\text{g mL}^{-1}$)

To prepare $100 \mu\text{g mL}^{-1}$, 10ml of the standard for each metal ions of stock solutions were pipetted and added in to 100 ml volumetric flask finally diluted to the mark with deionized water and the solution was mixed thoroughly.

2.4.7: L_1 Reagent Solution (1×10^{-3} M)

A6-((1E)-((2-(((E)-2-hydroxy-3-methoxy-4-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)benzylidene)amino)benzyl)imino)methyl)-2-methoxy-3-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)phenol(L_1) reagent solution at concentration of 1×10^{-3} M was prepared by dissolving 0.0193 gm into a 10 ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 25 ml volumetric flask, and then diluted to the mark with DMSO .

2.4.8: L_2 Reagent Solution (1×10^{-3} M)

A6,6'-((1E,1'E)-(1,2-phenylenebis(azanylylidene))bis(methanylylidene))bis(3-(phenyldiazenyl)phenol)(L_2) reagent solution at concentration of 1×10^{-3} M was prepared by dissolving 0.0131 gm into a 10 ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 25 ml volumetric flask, and then diluted to the mark with DMSO .

2.4.9: L_3 Reagent Solution (1×10^{-3} M)

A6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene))bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol)(L_3) reagent solution at concentration of 1×10^{-3} M was prepared by dissolving 0.0157 gm into a 10 ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 25 ml volumetric flask, and then diluted to the mark with DMSO .

2.4.10: L_4 Reagent Solution (1×10^{-3} M)

A6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene))bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol)(L_4)
reagent solution at concentration of 1×10^{-3} M was prepared by dissolving 0.0146 gm into a 10 ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 25 ml volumetric flask, and then diluted to the mark with DMSO .

2.4.11: Sodium Hydroxide (0.05M)Solution

This solution was prepared by accurately weighing 0.5g of material in into a 10 ml beaker, added minimum amount of deionized water and swirled to dissolve. The content was transferred quantitatively to a 250 ml volumetric flask, and then diluted to the mark with deionized water .

2.4.12: Hydrochloric Acid (0.05M) Solution

This solution was prepared by slowly adding with stirring 1.036ml of concentrated HCl ($d=1.19$ g/ml, 37% w/w) to appropriate amount of deionized water in 250 ml volumetric flask and dilute to mark with deionized water.

2.5: Primary testing of complex formation between metal ions and reagents

In a number of test tube added (1 ml) of metal solution ($100 \mu\text{g}.\text{ml}^{-1}$) then added (2ml) of reagent solution (1×10^{-3} M) drop wise with mixing and the change of color is observed divided the solution in two halve add few drops of 0.05 M of NaOH for one halve and 0.05M of HCl for another the show in table (2.4) .

Table (2.4): Primary test for reaction between metal and reagent in 0.05M NaOH but no there are changing in the color in present 0.05M HCl

No.	Ions	Ion solution color	Ion solution color after adding the reagent L ₁	Ion solution color after adding the reagent L ₂	Ion solution color after adding the reagent L ₃	Ion solution color after adding the reagent L ₄	The result
1	Cu ²⁺	Light blue	Reddish brown	Reddish orange	Light orange	No color change	Cu ²⁺ with L ₁ , L ₂ and L ₃
2	Co ²⁺	Light purple	No color change	Yellow orange	Reddish orange	No color change	Co ²⁺ with L ₂ and L ₃
3	Cr ³⁺	Light green	No color change	No color change	Brown to black	Light brown	Cr ³⁺ with L ₃ and L ₄
4	Ni ²⁺	Light green	Red to Orange	No color change	No color change	No color change	Ni ²⁺ with L ₁
5	Cd ²⁺	Colorless	Orange	No color change	No color change	Yellow orange	Cd ²⁺ with L ₁ and L ₄

2.6: Study of Spectroscopic Measurement for Reagents

UV-Vis spectra were obtained for ($1 \times 10^{-3} \text{M}$) of an DMSO solution for all reagents using (1cm) quartz cell in the wavelength (200-600) nm for reagent and (360-800) nm for complexes .

2.7: General Procedure for spectrophotometric determination of Cu, Co, Cr, Ni and Cd by L_1, L_2, L_3 and L_4 reagents

After determining the metal ions that react to reagents in table (2-4), can be determination λ_{max} , volumetric flask of capacity (10ml) has been taken and (1ml) of the ion concentration of solution ($100 \mu\text{gml}^{-1}$) was added, (2ml) of reagent ($1 \times 10^{-3} \text{M}$) added and complete the volume to the mark with deionized water , the blank solution was prepared in the same way except for the addition of the ion under study .

2.8: Optimization of the experimental conditions

2.8.1: Effect of reagent volume

Reagent (L_1) of various volumes (0.5-4ml) of concentration ($1 \times 10^{-3} \text{M}$) were taken in 10ml volumetric flask , 1.0 ml of ($100 \mu\text{gml}^{-1}$) Cu(II) solution added and the mixture solution was well shaken then the volume was completed to the mark with deionized water. The absorbance of metal-chelates was recorded at λ_{max} of copper complex against reagent blank prepared in a same way without Cu(II). The same method was repeated for Cd(II) and Ni(II) ions with L_1 and also for the other reagents with selected metal ions .

2.8.2: Effect of reaction time

The effect of time was studied in order to signify the stability of the complexes optimum volume of reagent L_1 concentration ($1 \times 10^{-3} M$) was taken in 10ml volumetric flask, 1.0ml of ($100 \mu g \cdot ml^{-1}$) Cu(II) solution added and the mixture solution was well shaken then the volume was completed to the mark with deionized water. The absorbance of metal-chelates was recorded at λ_{max} of copper complex at (0-50) min against reagent blank prepared in a same way without Cu(II). The same method was repeated for Cd(II) and Ni(II) ions with L_1 and also for the other reagents with selected metal ions.

2.8.3: Effect of pH

The influence of pH value on the absorbance of complexes was studied at different pH by using HCl and NaOH (0.05M) solutions at (pH 2-12), volumetric flask (10ml) was taken put (1ml) of copper solution ($100 \mu g \cdot ml^{-1}$) and to solution added optimum volume of reagent L_1 solution ($1 \times 10^{-3} M$) after adjusting the pH then complete the volume to the mark. After appropriate time the absorbance was measured at appropriate λ_{max} of copper complex against a reagent blank prepared in a similar way without ions. The same method was repeated for Cd(II) and Ni(II) ions with L_1 and also for the other reagents with selected metal ions.

2.8.4: Effect for temperature

The effect temperature was studied by using volumetric flask (10ml) were taken put (1ml) of copper solution ($100 \mu g \cdot ml^{-1}$) in volumetric flask (10ml) and added optimum volume of reagent L_1 ($1 \times 10^{-3} M$) after adjusting the pH then complete the volume to the mark, and put it in a water bath at different temperatures at (20 – 60) °C after appropriate

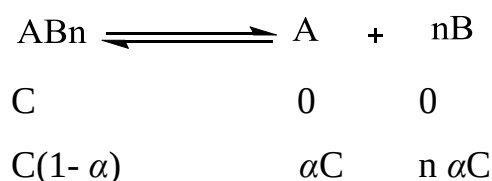
optimum time. The absorbance was measured at appropriate $\lambda_{\max}$ of complexes against a reagent blank prepared in a similar way without ions. The same method was repeated for Cd(II) and Ni(II) ions with L_1 and also for the other reagents with selected metal ions .

2.9:Determination of dissociation degree and stability constant

The stability constants are obtained by measuring the absorbance of solution mixture of ligand and metal ion at fixed wavelength ($\lambda_{\max}$) and optimum pH values. The conditional or apparent stability constant of (Metal : Ligand) product were evaluated as follows:

In order to measure A_m and A_s value used to calculate the dissociation degree and stability constant. This was done by taking equivalent volumes from the reagent and metal ion solutions ($1 \times 10^{-4} M$) and after optimum condition of each complex the absorbance was measured at appropriate $\lambda_{\max}$ against a reagent blank prepared in a similar way without ions, and we obtain (A_s) which represent the complex in partial dissociation. The experiment return by excess the volume of reagent then the absorbance was measured at appropriate $\lambda_{\max}$ of complexes against a reagent blank prepared in a similar way without ions to obtain (A_m) which represent the undissociation complex. The interaction between the metal ions and the reagents proceeds according to the equations⁽¹⁸⁴⁾ :

$$\alpha = \frac{A_m - A_s}{A_m}$$



$$(\alpha C)(n \alpha C)^n$$

$$K_{\text{inst}} = \frac{1}{C(1-\alpha)}$$

Where

C = concentration of the ligand which is equal the concentration of the metal.

α = degree of dissociation

A_s = the absorption of solution containing a stoichiometric amount of ligand and metal ion

A_m = the absorbance of solution containing the same amount of metal and excess of ligand.

n = no. of mole

The stability constant is equal to

$$K_{\text{st}} = \frac{1}{K_{\text{inst}}}$$

2.10: Determination of the stoichiometry of the complex

The stoichiometry of the complexes were investigated by Continuous variation method (Job's method) in order to find M:L ratio, the absorption of metal ion and ligand solution mixture was measured at λ_{max} .

Continuous variation method (Job method)

In this method, variable volumes of solutions containing same molar concentration ($1 \times 10^{-4} \text{M}$) of ligand and metal ions were mixed together. The total volume was kept constant at 2ml. The absorbance of each solution was measured at λ_{max} and the relation between the mole fraction of the volume $V_m / (V_m + V_L)$ on the X-axis (where V_m and V_L represent the volume of the metal and ligand solutions respectively) was plotted against the absorbance on the Y-axis.

2.11: Preparation of azo–azomethine complexes

The azo-Azomethine complexes were prepared by dissolving the ligand (L_2 , L_4) (0.0005mole) in 20ml hot ethanol and added dropwise with a stoichiometric amount of 1:1 [M:L] molar ratio (0.0005mole) for Co(II) and Cu(II) chloride salts with L_2 , Cr(II) and Cd(II) chloride salts with L_4 dissolved in 20ml hot pH solution from (0.05M HCl and 0.05M NaOH) at optimal pH for each metal ions. The reaction mixture was heated to (30-45) °C for 50 min, until solid complexes were precipitated then left overnight. The solid product thus formed was filtered off, washed distilled water until the solution become colourless and washed with 10ml hot ethanol to remove any traces of the unreacted materials and left to dry. The recrystallized using absolute ethanol⁽¹⁸⁵⁾.

The table (2.5): Some physical and chemical properties for the prepared ligand and their complexes

No.	Chemical formula	M.Wt	pH	Color	$\lambda_{\max}$	m.p °C	Yeild%
1	$C_{39}H_{32}N_8O_6S_2$ (L_1)	772	6	Light brown	283	288-290	80.37
2	$C_{32}H_{24}N_6O_2$ (L_2)	524	6	Brown	238	210-212	80.00
	CoL_2Cl_2	654	7	Dark brown	600	282-284	82.57
	CuL_2Cl_2	659	6	Black	647	261-263	80.55
3	$C_{37}H_{34}N_6O_4$ (L_3)	626	6	Reddish orange	290	253-254	78.13
4	$C_{34}H_{28}N_6O_4$ (L_4)	584	6	Dark brown	396	194-196	79.57
	CdL_4Cl_2	767	6	Orange	602	263-265	74.48
	CrL_4Cl_2	742	7	Dark orange	530	220-221	78.85

2.12: Preparation of Standard Solutions of metals and Reagents for cloud point extraction

2.12.1: L_1 Reagent Solution (1×10^{-2} M)

A6-((1E)-((2-(((E)-2-hydroxy-3-methoxy-4-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)benzylidene)amino)benzyl)imino)methyl)-2-methoxy-3-((6-methoxybenzo[d]thiazol-2-yl)diazenyl)phenol(L_1) reagent solution at concentration of 1×10^{-2} M was prepared by dissolving 0.7728 gm into a 50 ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 100ml volumetric flask, and then diluted to the mark with DMSO. Other standard solutions were prepared by dilution with the same solvent.

2.12.2: L_2 Reagent Solution (1×10^{-2} M)

A6,6'-((1E,1'E)-(1,2-phenylenebis(azanylylidene))bis(methanylylid ene))bis(3-(phenyldiazenyl)phenol)(L_2) reagent solution at concentration of 1×10^{-2} M was prepared by dissolving 0.5246gm into a 50ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 100ml volumetric flask, and then diluted to the mark with DMSO. Other standard solutions were prepared by dilution with the same solvent.

2.12.3: L_3 Reagent Solution (1×10^{-2} M)

A6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene))bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol)(L_3) reagent solution at concentration of 1×10^{-2} M was prepared by dissolving 0.6267gm into a 50ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 100ml

volumetric flask, and then diluted to the mark with DMSO. Other standard solutions were prepared by dilution with the same solvent .

2.12.4: L_4 Reagent Solution (1×10^{-2} M)

A6,6'-((1E,1'E)-((4-methyl-1,2-phenylene)bis(azanylylidene))bis(methanylylidene))bis(2-methoxy-3-(o-tolyldiazenyl)phenol)(L_4) reagent solution at concentration of 1×10^{-2} M was prepared by dissolving 0.5846 gm into a 50 ml beaker, added minimum amount of DMSO and swirled to dissolve. The content was transferred quantitatively to a 100 ml volumetric flask, and then diluted to the mark with DMSO. Other standard solutions were prepared by dilution with the same solvent .

2.12.5: Hydrochloric Acid (6M) Solution

This solution was prepared by slowly adding with stirring 49.75ml of concentrated HCl ($d=1.19$ g/ml, 37%) to appropriate amount of deionized water in 100ml volumetric flask and dilute to mark with deionized water. Other standard solutions were prepared by dilution with deionized water.

2.12.6: Nitric Acid (1%) Solution

This solution was prepared by slowly adding with stirring 1.45ml of concentrated HNO_3 to appropriate amount of deionized water in 100ml volumetric flask and dilute to mark with deionized water.

2.12.7: Nitric Acid Explained (65%) Solution

This solution was prepared by slowly adding with stirring 94.203ml of concentrated HNO_3 to appropriate amount of deionized water in 100 ml volumetric flask and dilute to mark with deionized water.

2.12.8: Hydrogen peroxide (30%) solution

This solution was prepared by adding 60ml of H_2O_2 in 100ml volumetric flask and dilute to mark with deionized water.

2.13: General Procedure of CPE⁽¹⁸⁶⁾

A typical cloud point experiment required the following steps: an aliquot of 10 ml of a solution containing a known of Cd(II) solution in 1ml of 0.1M HCl, 1ml of 1×10^{-3} M of reagent solution (L_1), 0.5 ml of 1% Triton X-100 were mixed. The mixture was shaken for 1min and left to stand for 20 min controlled by thermo-stated bath at 50°C until formation cloud point layer CPL. Separation of the two phases was achieved immediately where the surfactant-rich phase became a highly viscous (without need of centrifugation and cooling) and settled down at the bottom of the tube making the aqueous phase was easily discarded by decantation. After then, the surfactant rich phase in the tube was dissolved in 1ml of 1M HNO_3 in methanol and the absorbance of the resulting solution was measured at λ_{max} of Cd (280.28 nm) by FAAS for extracted ion complex and with used calibration curve was measured the concentration of extracted Cd(II) amount, For the determine remainder quantity of Cd(II) in aqueous solution after extraction by subtraction quantity of Cd(II) measured in the surfactant-rich phase from original quantity, afterward calculate Distribution ratio (D).

The same procedure was applied for Cu(II) or Co(II) ions with L_2 , L_3 respectively. Thus the distribution ratio (D) can be calculated from the following equation:

$$D = \frac{[\text{M}^{n+}]_{\text{cpL}}}{[\text{M}^{n+}]_{\text{aq}}} \dots\dots\dots(2-1)$$

where

$[M^{n+}]_{cpL}$: metal ion concentration that transferred to micelle phase

$[M^{n+}]_{aq}$: metal ion concentration that remained (residual) in aqueous phase.

2.14: Optimization of the Parameters for CPE

The effects of several experimental parameters which impact the CPE efficiency were carried out by classical optimization (OFAT) procedure. In as much as the extraction efficiency of the CPE depends on dual factors, some of regarding the prior formation of a complex with sufficient hydrophobicity and the other for the formation of micelles to obtain the desired separation and preconcentration. Consequently, the effects of reagent concentration, HCl concentration, incubation time, equilibration temperature and Triton X-100 volume, were selected in this study.

2.14.1: Effect of reagent Concentration

This study was carried out by taking variety concentration of reagent solution as complexing agent of (1×10^{-4} , 5×10^{-4} , 1×10^{-3} , 5×10^{-3} , 1×10^{-2} , $5 \times 10^{-2} M$) were added to 5ml of $3 \mu g \cdot ml^{-1}$ Cd(II), 1ml of 0.1 M HCl, 0.5ml of 1% Triton X-100 in 10 ml separated volumetric flasks and diluted to marks with deionized water and mixed thoroughly. The contents of each flask was subjected to general CPE procedure (2.13), in formation of ion-association complex with enough hydrophobicity. The same method was repeated for the reagent (L_2) with (Cu^{2+}) and (L_3) with (Co^{2+}).

2.14.2: Effect of HCl Concentration

In the separated 10mL volumetric flask containing 5ml of $3\mu\text{gml}^{-1}$ of Cd(II), 1ml of $5 \times 10^{-3}\text{M}$ of reagent solution(L_1), 0.5 ml of 1% Triton X-100 and (1ml) of varying concentration of HCl (0.05-2) M were mixed and diluted to marks with deionized water. The contents of each flask was subjected to general CPE procedure (2.13), in formation of their ion-association complex with enough hydrophobicity. The same method was repeated for the reagent (L_2) with (Cu^{2+}) and (L_3) with (Co^{2+}) .

2.14.3: Effect of the Incubation Time

The effect of incubation or heating time was examined on the extraction. It varied from (10 - 45) min for eight solutions of $3\mu\text{g.ml}^{-1}$ Cd (II), keeping the concentration of reagent constant and other parameters at optimum conditions subjected to the general CPE procedure (2.13). The result can be shown. The same method was repeated for the reagent (L_2) with (Cu^{2+}) and (L_3) with (Co^{2+}).

2.14.4: Effect of Equilibrium Temperature

A series of individual experiments were conducted to study the effect equilibrium temperature on the extraction of $3\mu\text{gml}^{-1}$ of Cd(II) keeping the concentration of reagent constant and other parameters at optimum conditions. The temperature was varied from 30 °C to 90 °C in a search of optimum values. The content of each flask was subjected to general CPE procedure (2.13) to find the distribution ratio (D) at each temperature setting. The result can be shown in figure (3.65). Thereafter the extraction constant K_{ex} and thermodynamic parameters were calculated according to equations (2-2) to (2-6) to predict the cases of solubilization of ion-pair complexes into micelles by cloud point

methodology (CPE)⁽²¹⁰⁾. The same method was repeated for the reagent (L₂) with (Cu²⁺) and (L₃) with (Co²⁺) .

$$K_{ex} = \frac{D}{[M^{n+}]_{aq} [R]_s} \dots\dots\dots(2-2)$$

Where D is distribution ratio, [Mⁿ⁺] aq metal ion concentration that remained in aqueous phase and [R]_s concentration of reagent in surfactant phase.

After drawing a log K_{ex} value vs. 1/T °K , a straight line relationship is obtained and from a slope value the thermodynamic parameters (ΔH_{ex} , ΔG_{ex} , ΔS_{ex}) were calculated for extraction process from following equations :

Vant-Hoff equation to calculate the thermodynamic value ΔH_{ex}⁽¹⁸⁷⁾

$$\frac{\Delta \log K_{ex}}{\Delta 1/T} = \frac{-\Delta H_{ex}}{2.303 R} \dots\dots\dots (2-3)$$

$$slope = \frac{-\Delta H}{2.303 R} \dots\dots\dots (2-4)$$

$$\Delta G_{ex} = -RT \ln K_{ex} \dots\dots\dots (2-5)$$

$$\Delta G_{ex} = \Delta H_{ex} - T\Delta S_{ex} \dots\dots\dots (2-6)$$

2.14.5: Effect Triton X-100 Volume

Different volumes of 1% Triton (X-100) ranging from (0.1-1.5) ml were added to 5ml of $3\mu\text{gml}^{-1}$ of Cd(II) ion solution, and present 1ml of 0.5M HCl and 1ml of 5×10^{-3} M of reagent solution(L_1) in 10 ml separated volumetric flasks and diluted to marks with deionized water and mixed thoroughly. The content of each flask was subjected to general CPE procedure (2.13). The same method was repeated for the reagent (L_2) with (Cu^{2+}) and (L_3) with (Co^{2+}) .

2.15: Application of real samples

The present CPE method was applied for determination of metal ions in tap, river waters, liquid milk, tobacco , black tea and soil. River water was collected from Al-Refaie / Thi-Qar and tap water were taken from laboratory. liquid milk , tobacco and black tea samples were taken from submarket .

2.15.1: Water sample Preparation

Water samples including (tap and river water) , to remove suspended particulate matter, river water sample was filtered through Watman No. 42 and stored in polyethylene bottle . 15 ml of it was treated according to the given CPE procedure (2.13), under optimum conditions. The levels of analyte ions in the samples were determined by flame atomic absorption spectrometry⁽¹²⁶⁾.

2.15.2: Digestion of samples**2.15.2.1: Tobacco sample preparation**

The cigarette tobacco was prepared by removing the papers and filters from 60 cigarettes of 3 packs. The samples were dried in an oven at a temperature of 80 °C for 6 hours. The dried tobacco was ground in a mortar with a pestle until powdered finely as much as possible for homogenization, to simplify weighing and to facilitate organic matter destruction (digestion)⁽¹⁸⁸⁾.

2.15.2.2: Tobacco digestion

The digestion procedure of Campbell and Plank was used 1g of dried, ground, and homogenized cigarette tobacco was transferred to crucible . 5.0 ml of concentrated HNO₃ acid was added and the crucible was covered with watch glass and allowed to stand overnight. The covered crucible was placed in a muffle furnace with temperature controller and heated at 200°C for 30 min. The flask was removed and allowed to cool and 2 ml of 30% H₂O₂ was added and digested at the same temperature and time in the same way. This step was repeated to ensure complete digestion. The digest was allowed to dry to 1 to 2ml at 150 °C. Then 5.0 ml of 1% HNO₃ was added to digest residue and filtered quantitatively through Whatman filter paper No.40 into a 25ml volumetric flask and made up to volume with deionized water⁽¹⁸⁸⁾. 15ml of it was treated according to the given CPE procedure (2.13), under optimum conditions .

This was subsequently analyzed for Cd(II), Cu(II) and Co(II) using flame atomic absorption spectrophotometer .

2.15.3: Raw cow's milk sample digestion

The concentrations in sample were analyzed according to the procedure outlined by Peters⁽¹⁸⁹⁾. The milk sample need to be brought in a clear solution for analysis by atomic absorption spectroscopy. For this reason the sample first digested to dissolve the sample and to remove the fat. 1gm of raw cow's milk is treated with 5 ml (65% nitric acid) and 2ml (30% hydrogen peroxide) and then digested on electric hot plate at 90°C and the temperature of this mixture was gradually increased to 120°C. Until brown fumes appeared, indicating completion of oxidation of organic matter. The organic matrix of milk was destroyed and left the elements in to clear solution, after cooling the clear solution was filtered into 25ml volumetric flask and completed to the mark with deionized water^(189,190). 15ml of it was treated according to the given CPE procedure (2.13), under optimum conditions.

2.15.4: Tea sample digestion

Accurately weighed (1g) of tea sample was transferred into a crucible and kept in a muffle furnace for ashing at 450° C for 3 hours and then 5ml of 6M HCl was added to the crucible. Further, the crucible containing acid solution was kept on a hot plate and digested to obtain a clean solution. The final residue was dissolved in 0.1M HNO₃ solution and made up to 25ml. 15ml of it was treated according to the given CPE procedure (2.13), under optimum conditions⁽¹⁹¹⁾.

2.15.5: Soil sample digestion

1.0 g of the soil sample was weighed into a 50 ml beaker and 9 ml of 65% HNO₃, 4 ml of 50% (v/v) HF, 1 ml of 37% v/v HCl and 2 ml of 75% v/v HClO₄ were then added and digested at 135°C for 2 h. The digest was slowly evaporated until the white fumes of HClO₄ appeared. The digest was filtered and made up to 25 ml with deionized water⁽¹⁹²⁾.

15ml of it was treated according to the given CPE procedure (2.13), under optimum conditions .

2.16: Slope Analysis for Stoichiometric Determination of Complex

The method involved the extraction of Cd^{2+} , Cu^{2+} and Co^{2+} ions as from 10 ml aqueous solution containing constant amount of $3\mu\text{gml}^{-1}$ of metal ion, 1ml of 0.1M of HCl, 0.5 ml of Triton X-100 and increasing reagent concentrations of ranging from $(5 \times 10^{-4} - 1 \times 10^{-2} \text{M})$. The experiments were subjected to the optimum conditions established by general CPE procedure (2.13). After separation, the surfactant-rich phase which contains ion-pair complex at each concentration ratio was diluted with 1ml of in 1M HNO_3 in methanol and the absorbance of each solution was measured at wavelength absorption maxima by FAAS for extracted ion complex and with used calibration curve was measured the concentration of extracted metal ion amount, for the determine remainder quantity of metal ion in aqueous solution after extraction by subtraction remainder from original quantity , afterward calculate Distribution ratio (D) and that data were plotted as a log D against log [L] to find the stoichiometry of the ion-pair complex formed.

Chapter Three

Results

and

Discussion

Results and Discussion

3.1: Ultra violet-Visible Spectra (determination of λ_{max})

The UV- visible spectrum of the synthesized of the azo- azomethine dyes that dissolved in DMSO were performed at room temperature in the range (200 - 600) nm and their complexes in the range (360 - 800) nm. The azo-azomethine dyes were composed of two main maximum absorption peaks that are shown in the figures (L_1 - L_4) due to free reagents. The first at wavelength in the range (238 - 325) nm due to transitions of ($\pi \rightarrow \pi^*$) in the aromatic ring⁽¹⁹³⁾ and the second absorption maximum has emerged at the wavelength λ_{max} in the range (400 - 470) nm due to the transition ($n \rightarrow \pi^*$)⁽¹⁹⁴⁾ which belongs to the bond (-C=N-) and (-N=N-) .

Spectrum UV – Vis shows absorption bands of prepared complexes within studies range, the ($\pi \rightarrow \pi^*$) and ($n \rightarrow \pi^*$) transitions observed in the spectrum of reagents were shifted to longer wavelength (bathochromic effect) values along which confirm the chelation of the reagent with metal ions⁽¹⁹⁴⁾. The absorption maximum λ_{max} of each complexes show in the range (530 - 665) nm and these packages are attributable to the d-d transitions of type $M \rightarrow L$ between the metal and ligand which confirms formation of the complexes⁽¹⁹⁵⁾.

In the complexes, ($\pi \rightarrow \pi^*$) transitions were shifted to longer wavelengths as a consequence of coordination when binding with a metal, confirming the imine nitrogen was coordinated to the metal atom⁽¹⁸³⁾. Its shift to longer wavelengths can be related to the donation of the lone pairs of the nitrogen atom of imine group to the metal ion⁽¹⁸³⁾. In

the metal complexes, new bands at higher wave lengths support the formation of strong (M–O) and (M–N) bonds⁽¹⁹⁶⁾.

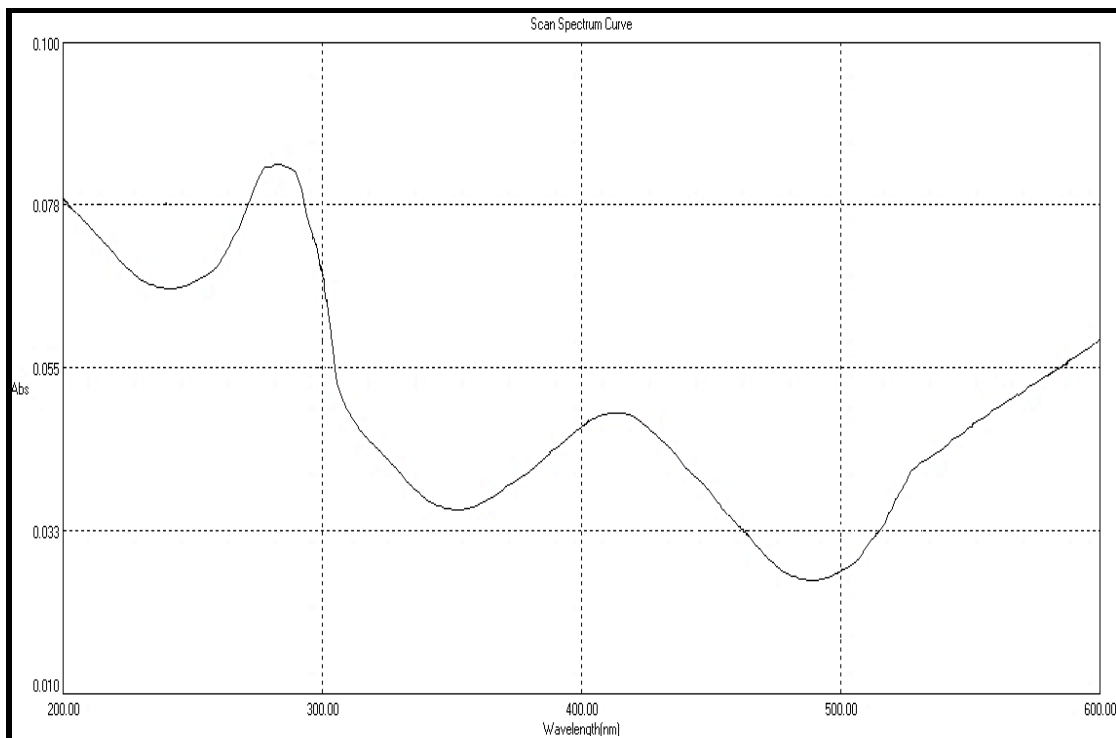


Figure (3.1): UV-Visible spectrum of L₁

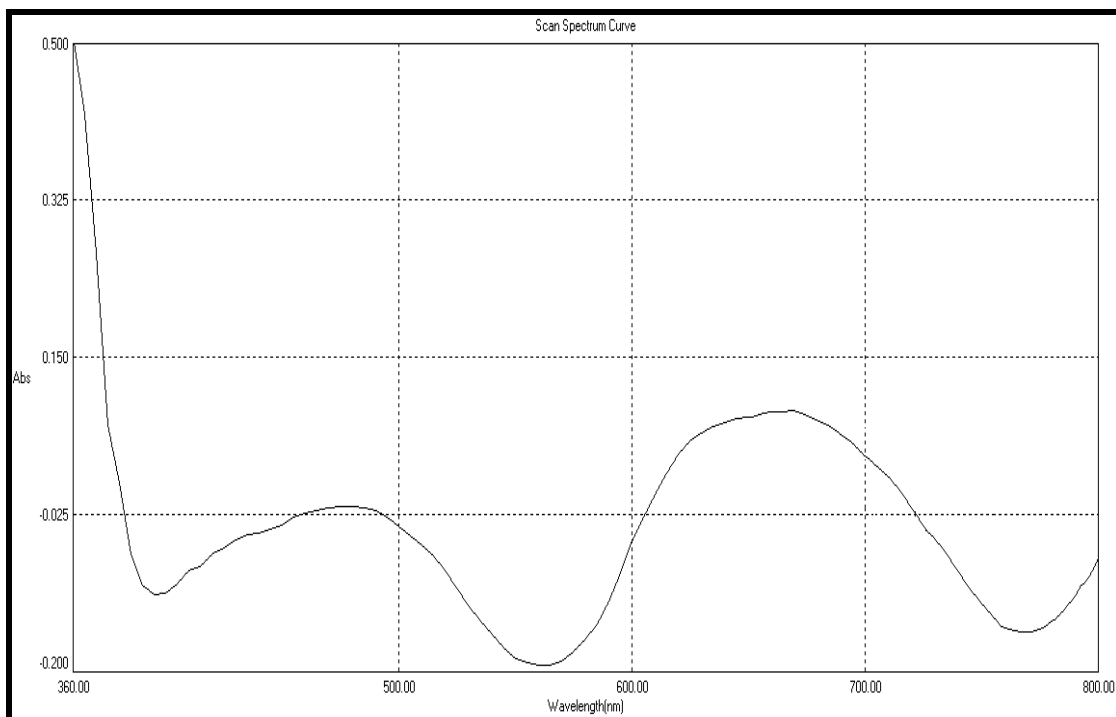


Figure (3.2): UV-Visible spectrum of [CuL₁Cl₂]

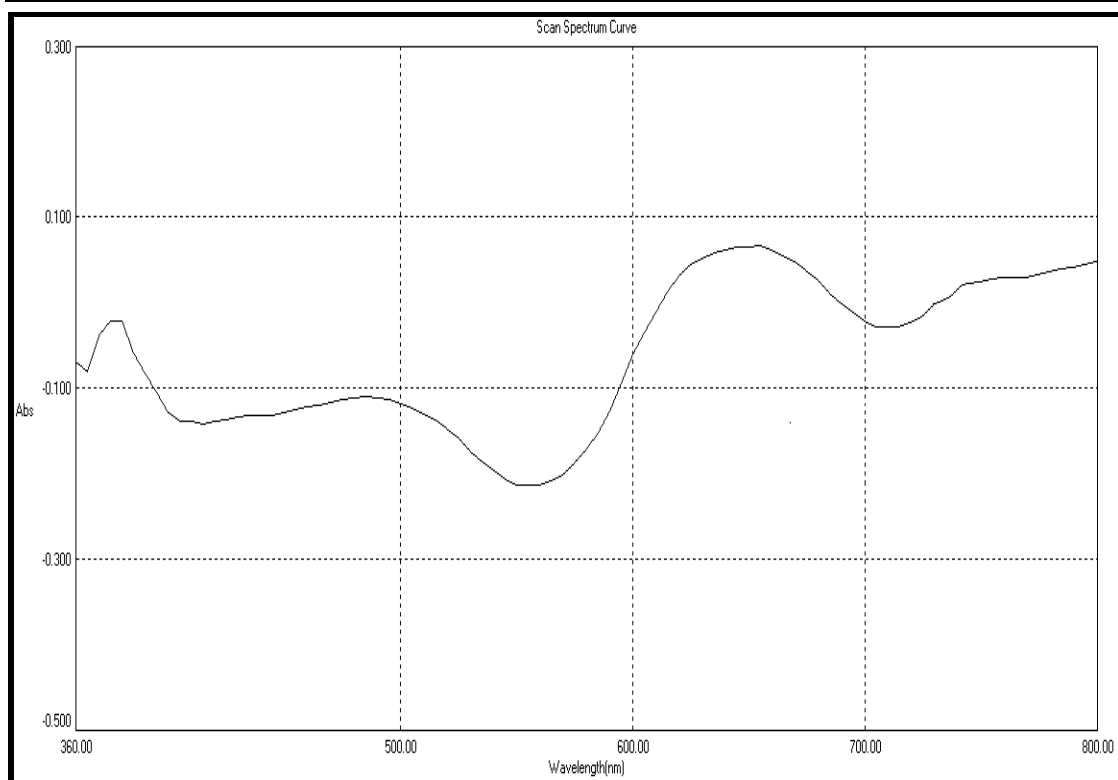


Figure (3.3): UV-Visible spectrum of $[\text{CdL}_1\text{Cl}_2]$

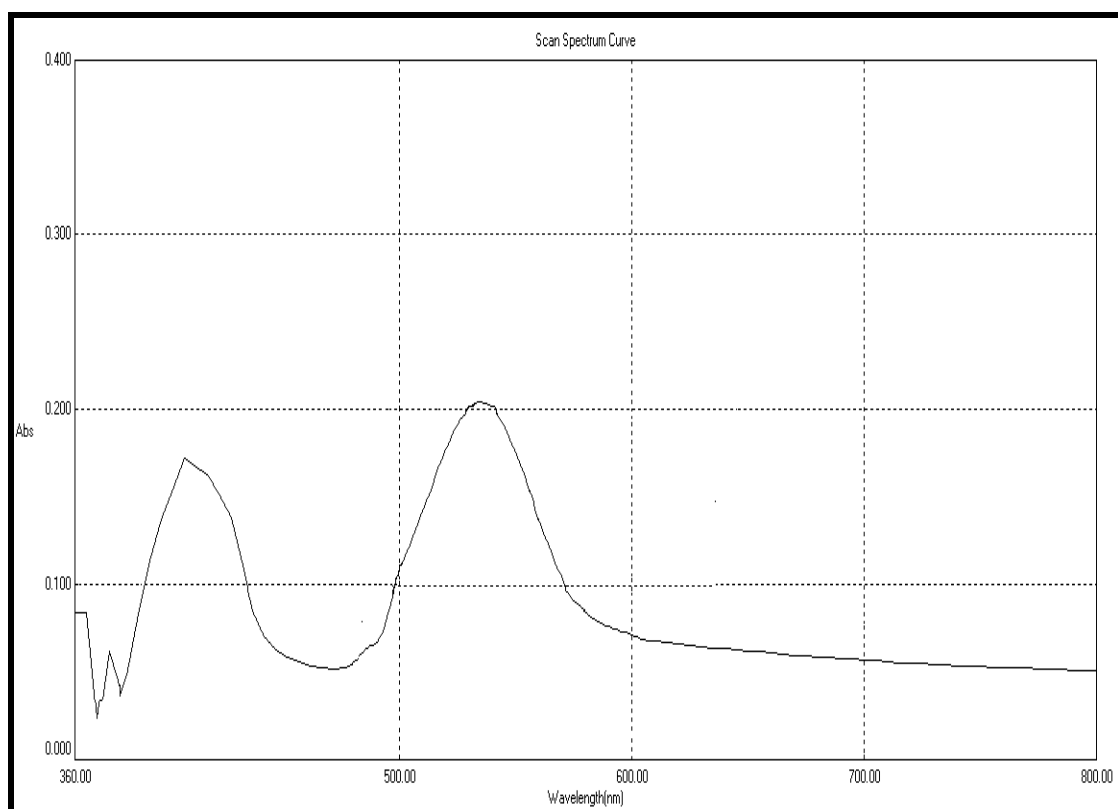


Figure (3.4): UV-Visible spectrum of $[\text{NiL}_1\text{Cl}_2]$

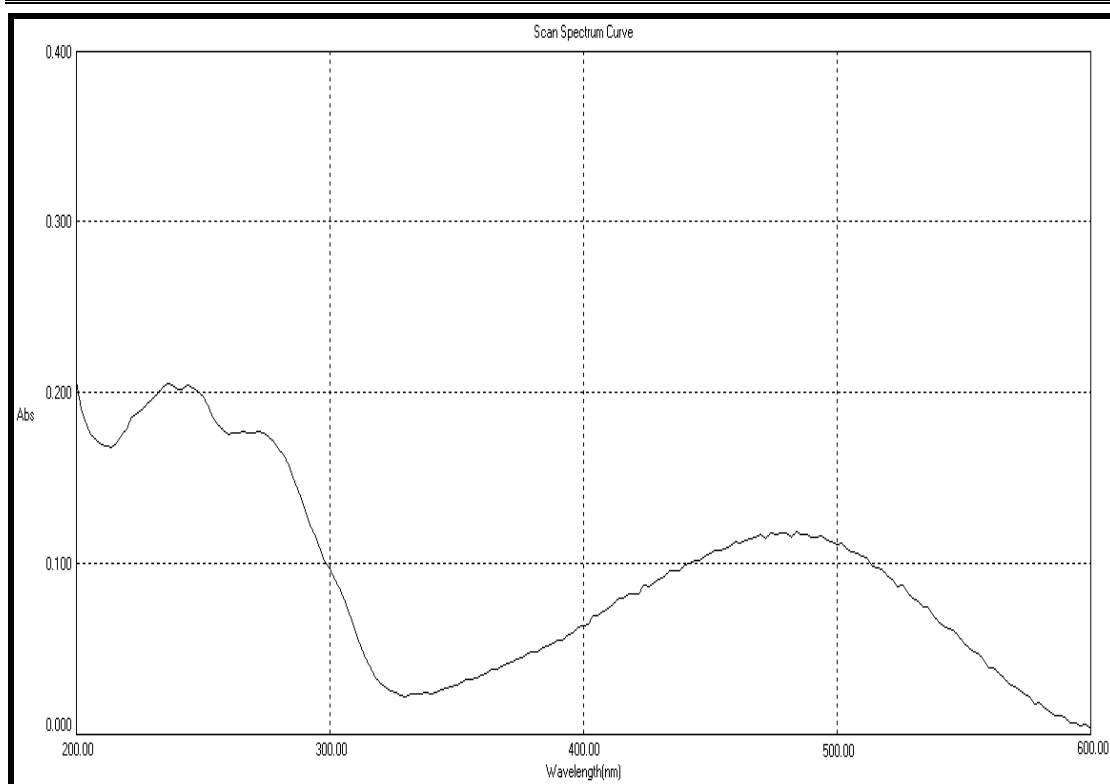


Figure (3.5): UV-Visible spectrum of L_2

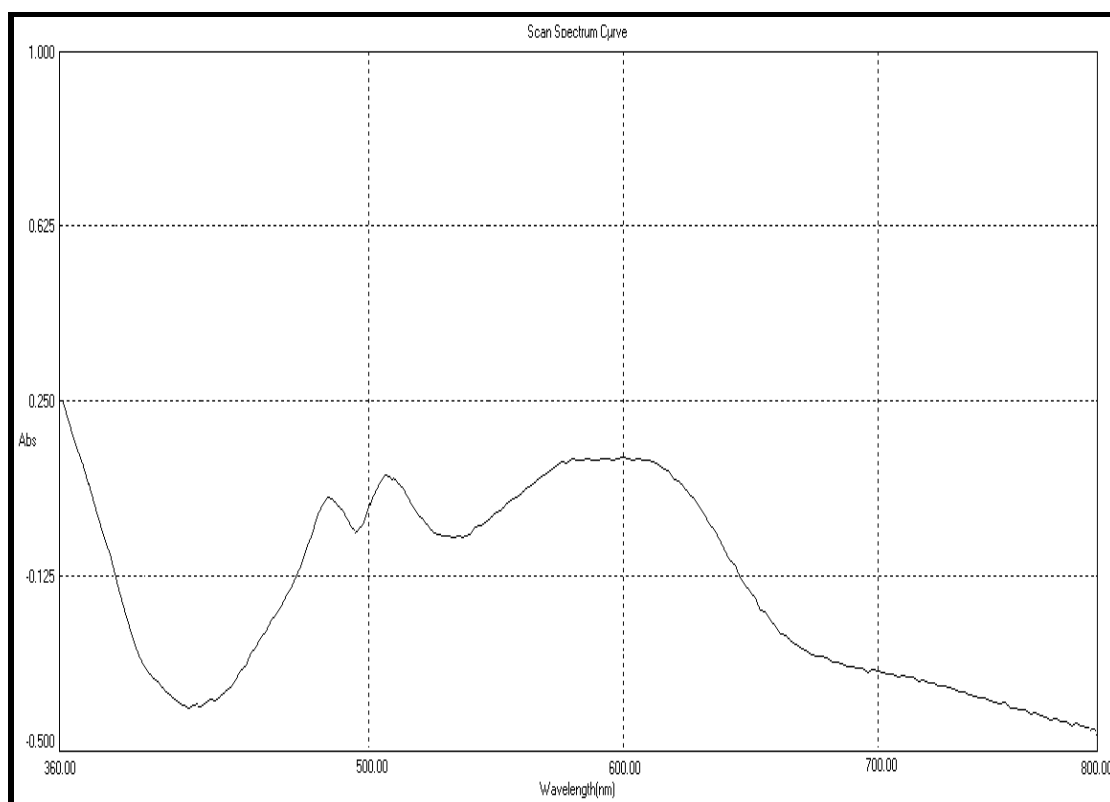


Figure (3.6): UV-Visible spectrum of $[CoL_2Cl_2]$

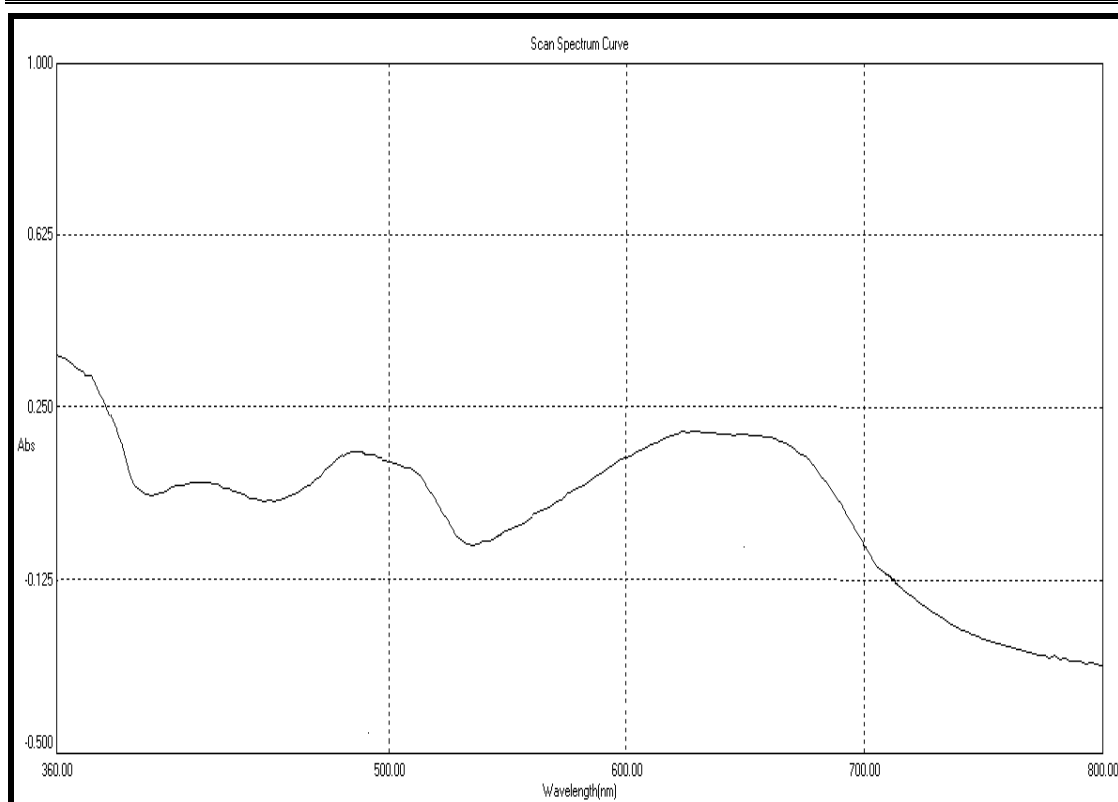


Figure (3.7): UV-Visible spectrum of $[\text{CuL}_2\text{Cl}_2]$

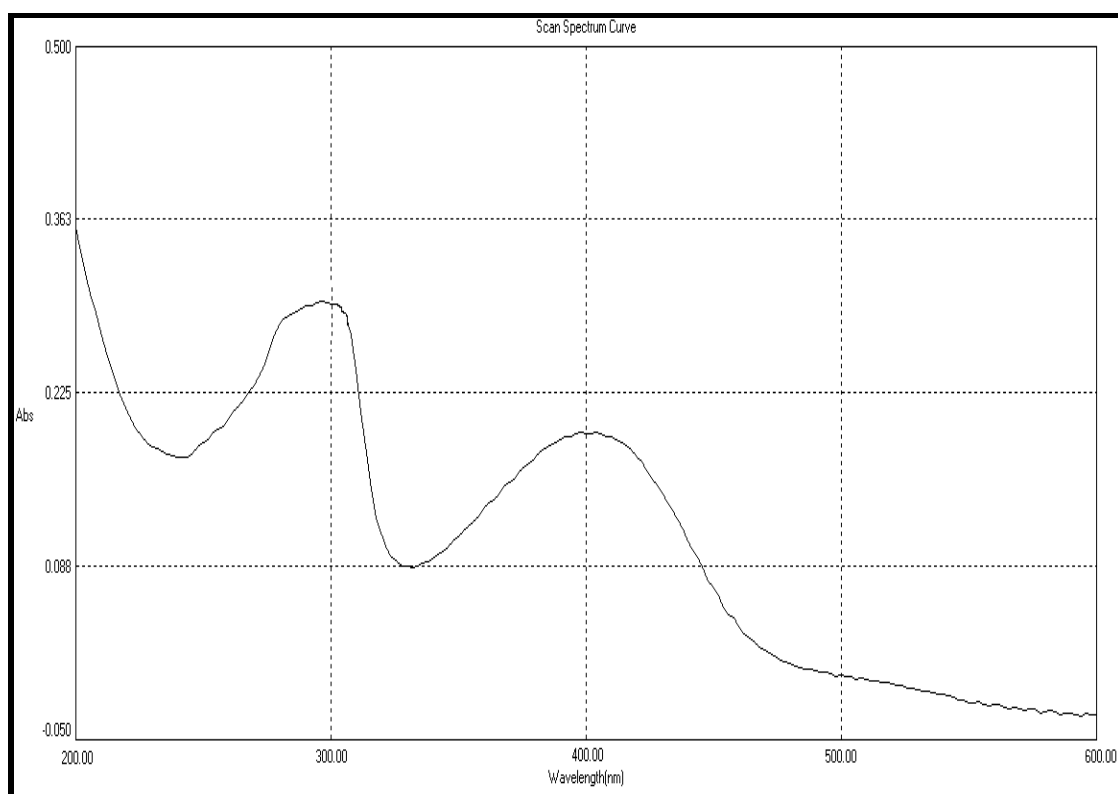


Figure (3.8): UV-Visible spectrum of L_3

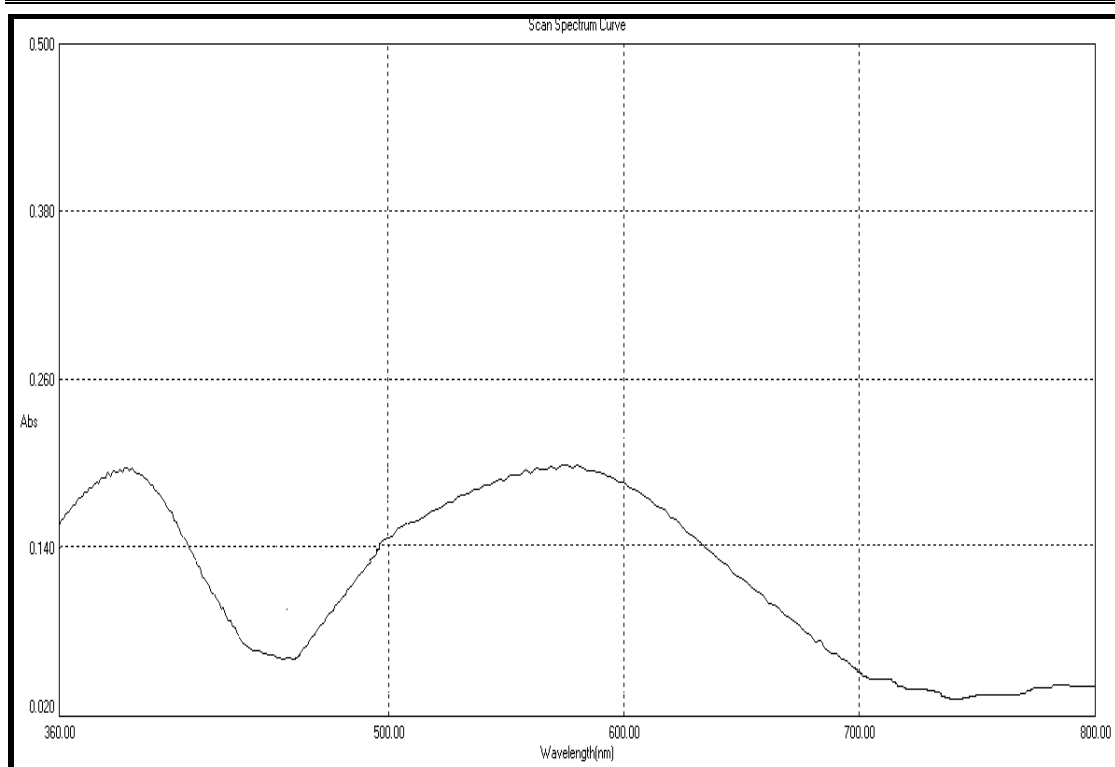


Figure (3.9): UV-Visible spectrum of $[\text{CuL}_3\text{Cl}_2]$

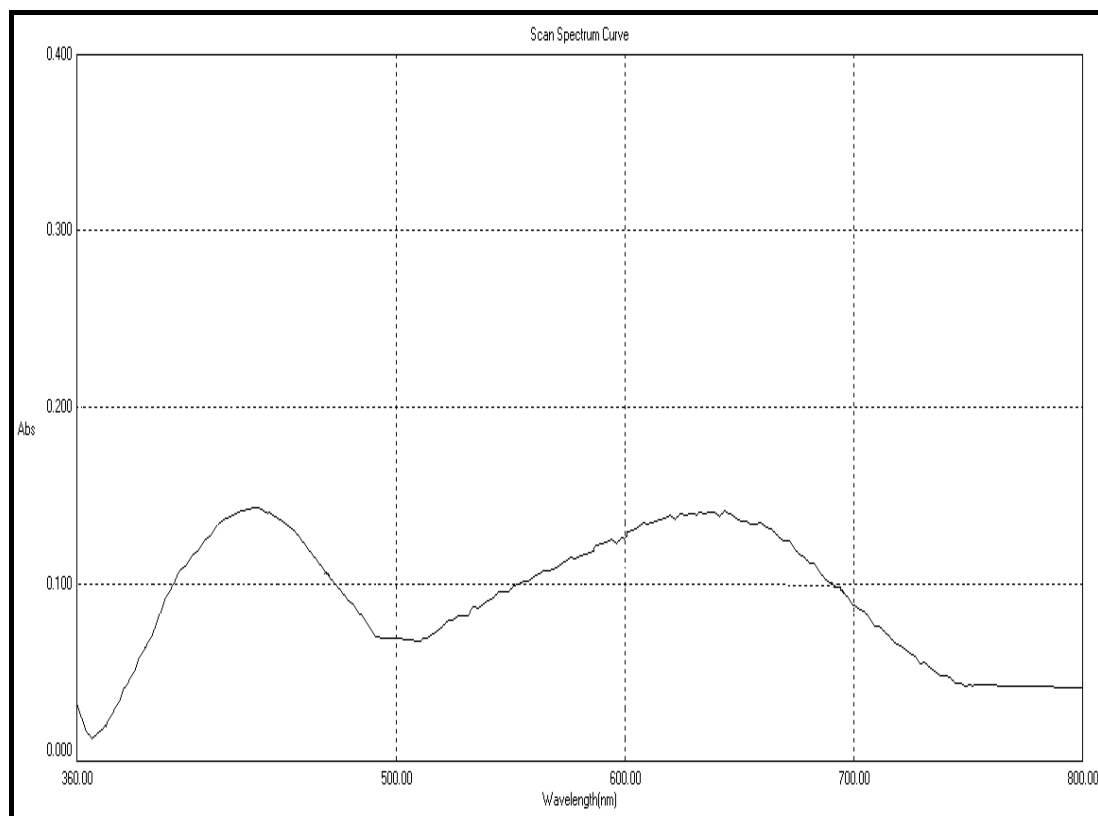


Figure (3.10): UV-Visible spectrum of $[\text{CrL}_3\text{Cl}_2]\text{Cl}$

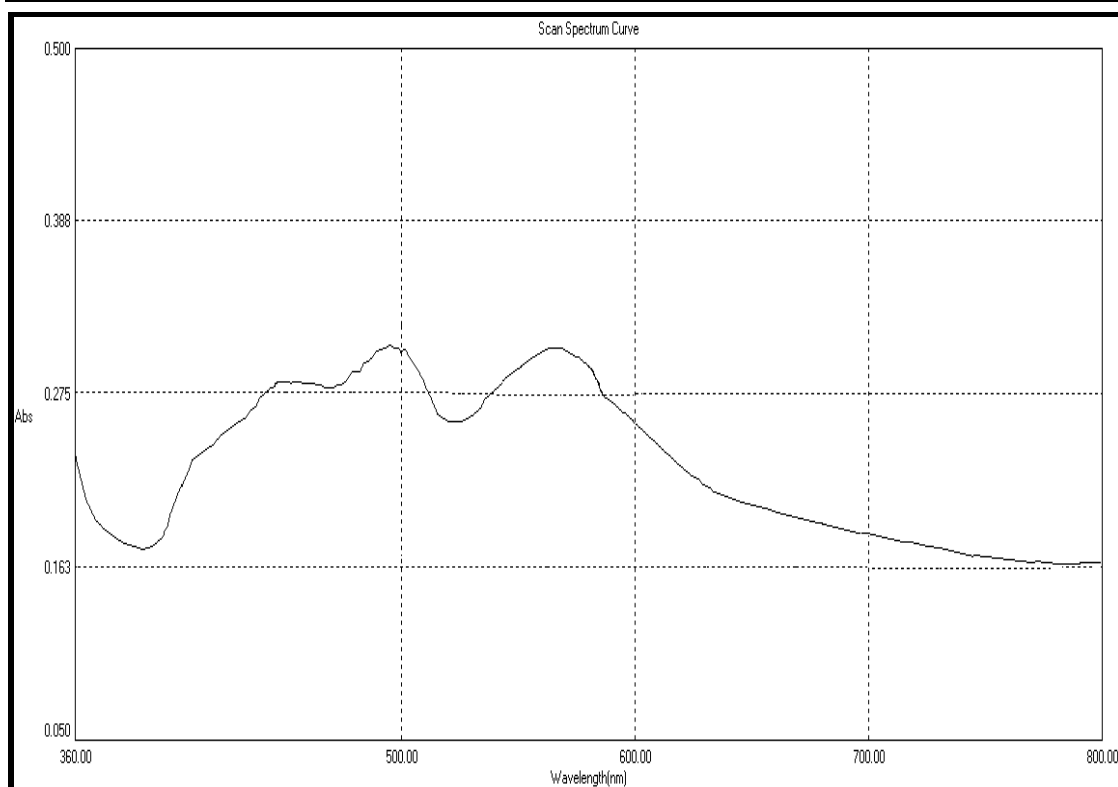


Figure (3.11): UV-Visible spectrum of $[\text{CoL}_3\text{Cl}_2]$

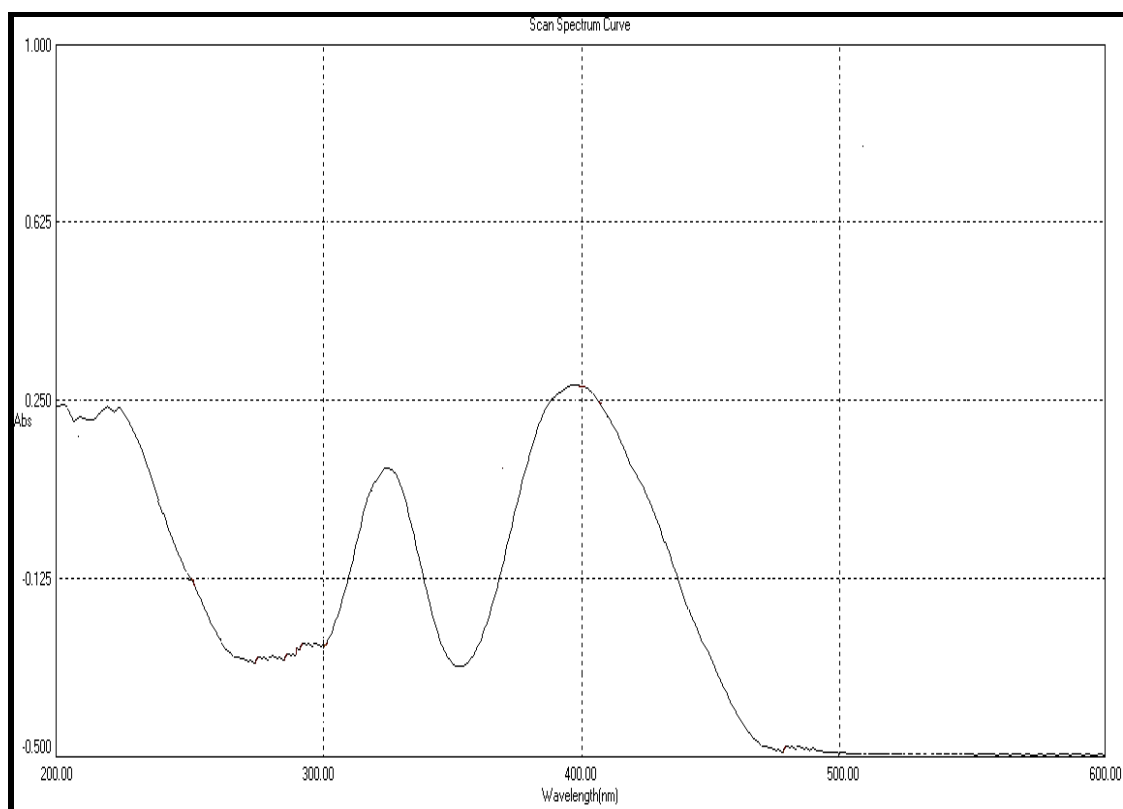


Figure (3.12): UV-Visible spectrum of L_4

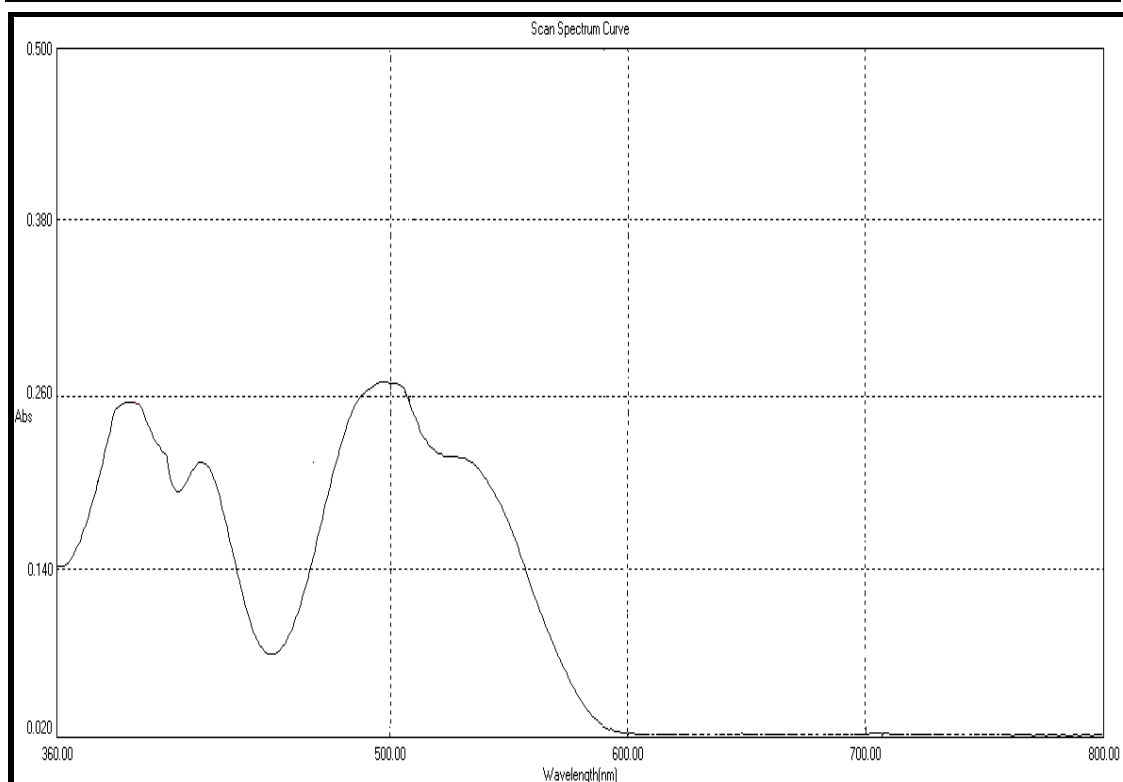


Figure (3.13): UV-Visible spectrum of $[\text{CrL}_4\text{Cl}_2]\text{Cl}$

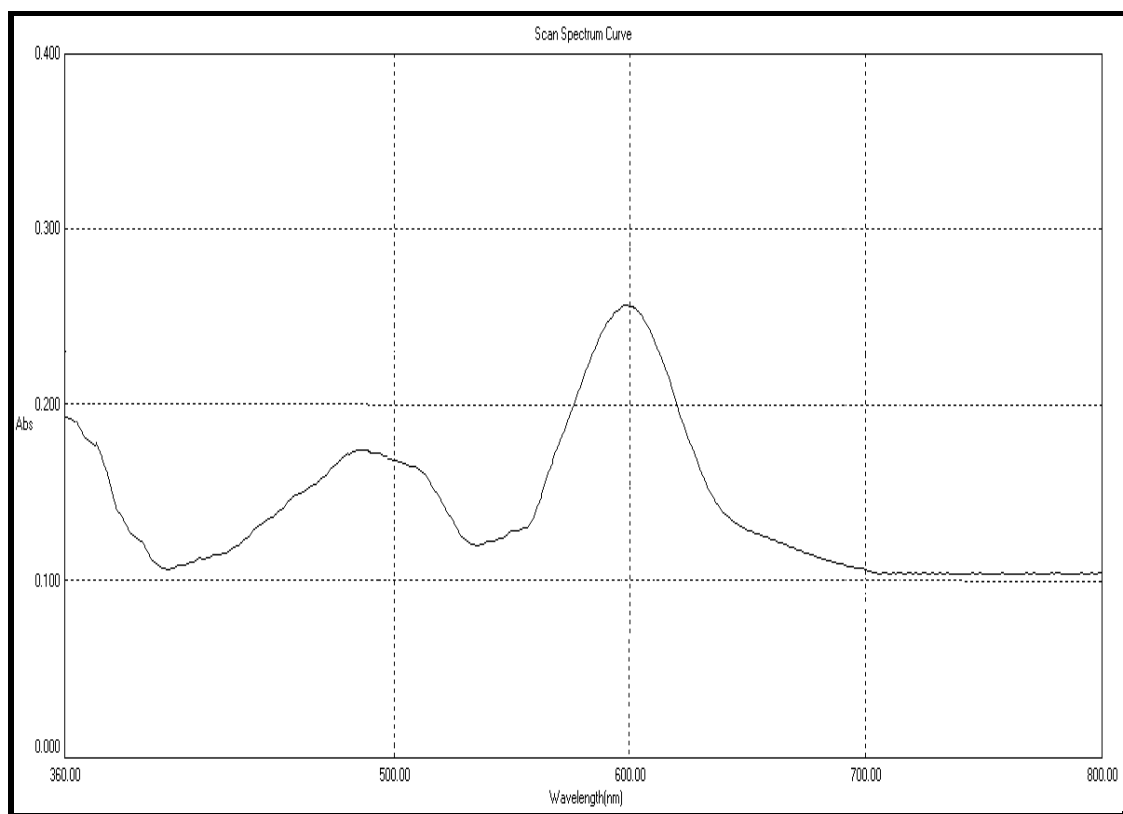


Figure (3.14): UV-Visible spectrum of $[\text{CdL}_4\text{Cl}_2]$

3.2: Optimization of the experimental Condition

3.2.1: Effect of reagent volume

The effect of reagent volume was studied by using different volumes (0.5–4.0) ml of the reagent ($1 \times 10^{-3} \text{M}$) with 1ml of $100 \mu\text{gml}^{-1}$ of metal and recorded the absorbance at λ_{max} . Highest absorbance at the 1ml of the reagent with Cd(II), 1.5ml of the reagent with Cu(II), 2ml of the reagent with Co(II), Ni(II) and Cr(III). It is optimized volumes for the formation of the complexes with metal ions. The results are shown in figures (3.15) - (3.18).

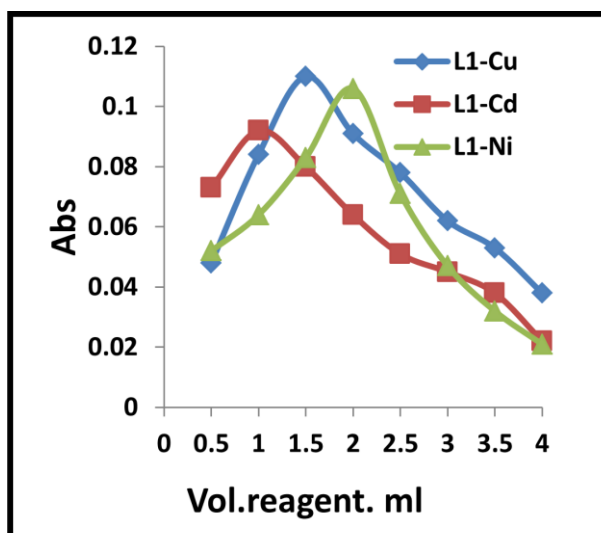


Figure (3.15): Effect of reagent volume at $\lambda_{\text{max}}=665$ nm for $\text{L}_1\text{-Cu}$, $\lambda_{\text{max}}=650$ nm for $\text{L}_1\text{-Cd}$ and $\lambda_{\text{max}} = 540$ nm $\text{L}_1\text{-Ni}$ complexes

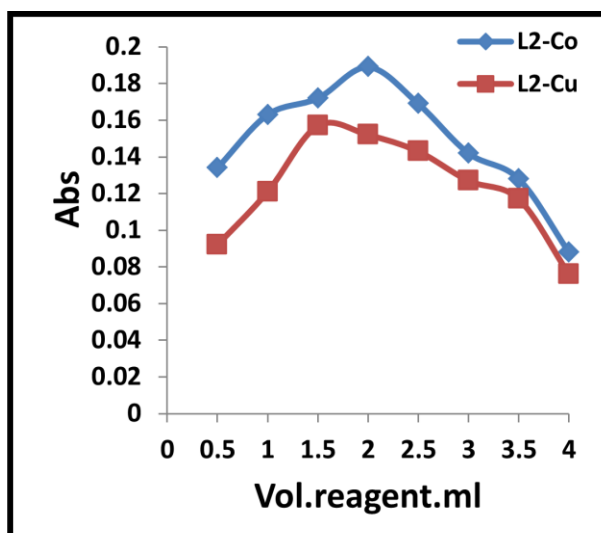


Figure (3.16): Effect of reagent volume at $\lambda_{\text{max}}=600$ nm for $\text{L}_2\text{-Co}$ and $\lambda_{\text{max}}=647$ nm for $\text{L}_2\text{-Cu}$ complexes

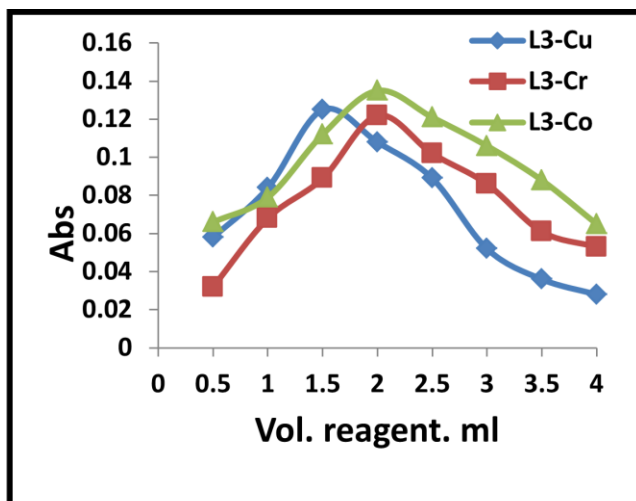


Figure (3.17): Effect of reagent volume at $\lambda_{\text{max}}=573$ nm for $L_3\text{-Cu}$, $\lambda_{\text{max}}=634\text{nm}$ for $L_3\text{-Cr}$ and $\lambda_{\text{max}}=566\text{nm}$ for $L_3\text{-Co}$ complexes

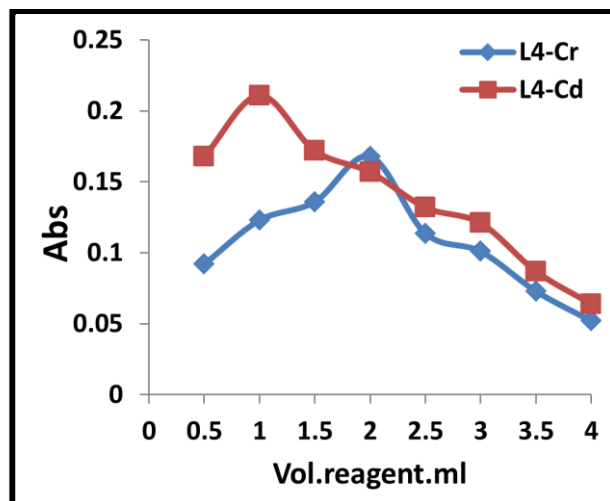


Figure (3.18): Effect of reagent volume at $\lambda_{\text{max}}=530$ nm for $L_4\text{-Cr}$ and $\lambda_{\text{max}}=602$ nm for $L_4\text{-Cd}$ complexes

3.2.2: Effect of reaction time

The effect of time to get the complex is an important factor to obtain knowledge of the time period which maintains the complex on stability that is especially employed in the field of analytical chemistry. The study was obtained at time (0-50) min. When the components were mixed at room temperature, maximum absorbance of the complexes were reached from (10-15) min from the moment start mixing and this useful in the field of spectral determination of metal ion. The reaction remains stable for 24 hr. This show the ligands strong coordination with metal ions⁽¹⁸⁵⁾. The results are shown in figures (3.19) – (3.22).

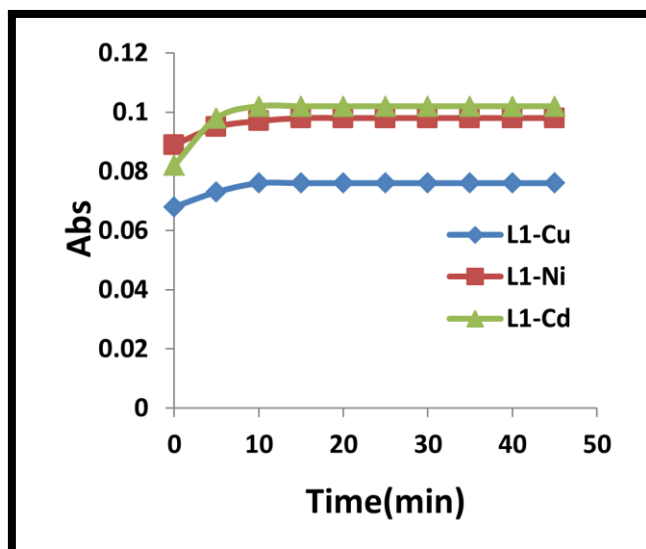


Figure (3.19): Effect of reaction time at $\lambda_{\text{max}}=665\text{nm}$ for $L_1\text{-Cu}$, $\lambda_{\text{max}} 540 \text{ nm}$ for $L_1\text{-Ni}$ and $\lambda_{\text{max}}=650\text{nm}$ for $L_1\text{-Cd}$ complexes

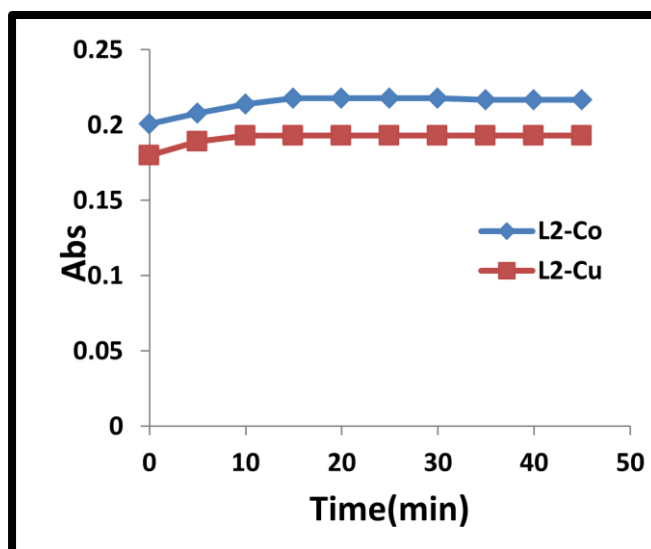


Figure (3.20): Effect of reaction time at $\lambda_{\text{max}}=600\text{nm}$ for $L_2\text{-Co}$ and $\lambda_{\text{max}}=647\text{nm}$ for $L_2\text{-Cu}$ complexes

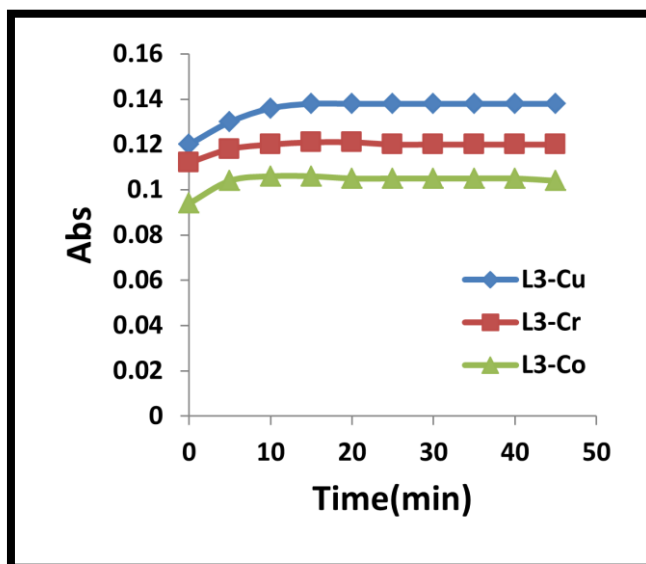


Figure (3.21): Effect of reaction time at $\lambda_{\text{max}}=573\text{nm}$ for $L_3\text{-Cu}$, $\lambda_{\text{max}}566 \text{ nm}$ for $L_3\text{-Co}$ and $\lambda_{\text{max}}=634\text{nm}$ for $L_3\text{-Cr}$ complexes

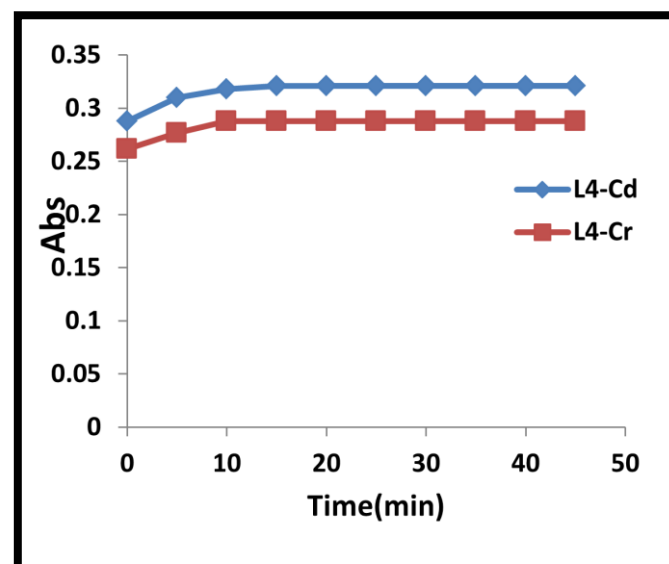


Figure (3.22): Effect of reaction time at $\lambda_{\text{max}}=530 \text{ nm}$ for $L_4\text{-Cr}$ and $\lambda_{\text{max}}=602 \text{ nm}$ for $L_4\text{-Cd}$ complexes

3.2.3: Effect of temperature

The effect of temperature on the absorbance of complex was examined. The study was performed at temperature between (20-60)°C. Figure(3.23)-(3.26) demonstrated that the maximum absorbance obtained at temperature range (30-45)°C which was viewed as an appropriate temperature of complex formation⁽¹⁹⁷⁾. At temperatures higher than 50°C the absorbance decreases due to dissociation of complex gradually⁽¹⁹⁸⁾.

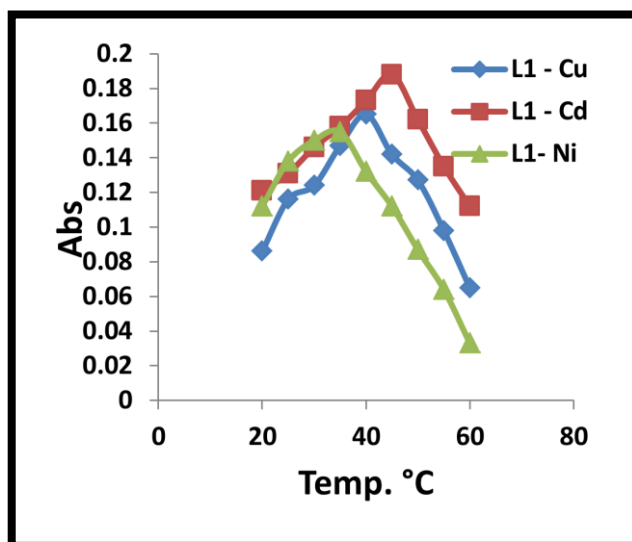


Figure (3.23): Effect of temperature at $\lambda_{\text{max}}=665\text{nm}$ for $L_1 - \text{Cu}$, $\lambda_{\text{max}}=540\text{ nm}$ for $L_1 -\text{Ni}$ and $\lambda_{\text{max}}= 650\text{ nm}$ for $L_1 - \text{Cd}$ complexes

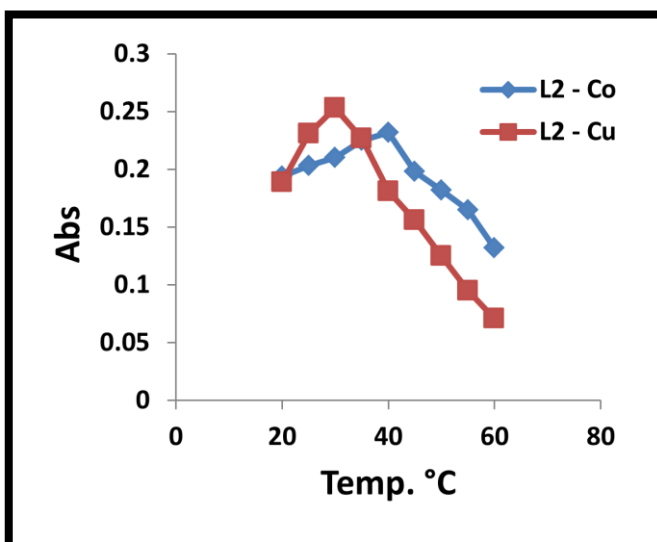


Figure (3.24): Effect of temperature at $\lambda_{\text{max}}=600\text{nm}$ for $L_2 - \text{Co}$ and $\lambda_{\text{max}}=647\text{nm}$ for $L_2 - \text{Cu}$ complexes

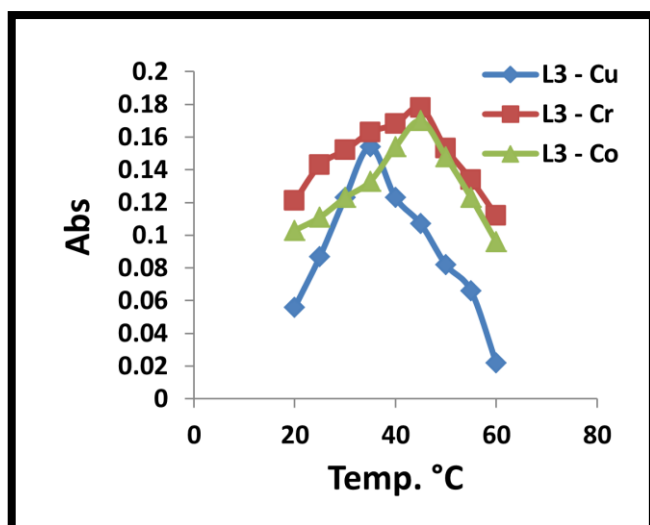


Figure (3.25): Effect of temperature at $\lambda_{\text{max}}=573\text{nm}$ for $L_3 - \text{Cu}$, $\lambda_{\text{max}} = 566\text{nm}$ for $L_3 - \text{Co}$ and $\lambda_{\text{max}} = 634\text{nm}$ for $L_3 - \text{Cr}$ complexes

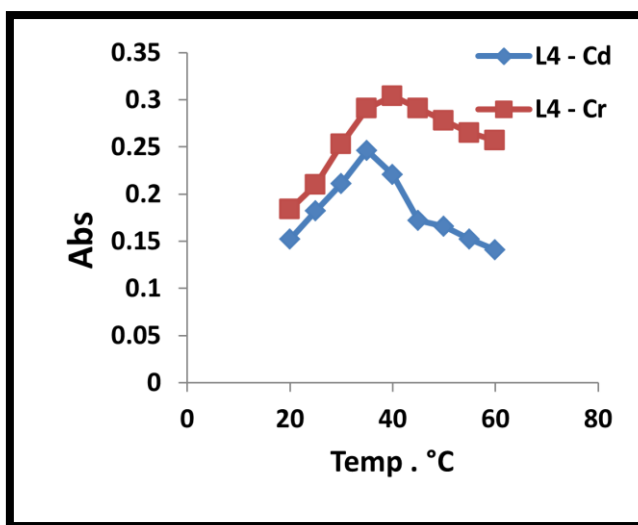


Figure (3.26): Effect of temperature at $\lambda_{\text{max}}= 530\text{ nm}$ for $L_4 - \text{Cr}$ and $\lambda_{\text{max}} = 602\text{ nm}$ for $L_4 - \text{Cd}$ complexes

3.2.4: Effect of pH

Figures (3.27) - (3.30) show that the absorbance of copper(II), nickel(II), cobalt(II), chromium(III) and cadmium(II) complexes measured at λ_{max} change with solution pH. The absorbance of each complex measured at λ_{max} starts low on the acidic side, reaches a maximum at the optimum pH value and then decreases again as pH is increased. The optimum pH values obtained from the plots give pH = 5 for Cu(II)⁽¹⁹⁹⁾ with L₁ and pH = 6 for Cu(II)⁽²⁰⁰⁾ with L₂ and L₃, pH = 8 for Ni (II)⁽¹⁹⁹⁾ with L₁, pH = 7 for Co(II)⁽¹⁹⁹⁾ with L₂ and pH = 8 for Co with(II)⁽¹⁸⁴⁾ with L₃, pH = 7 for Cr(III)⁽¹⁹⁷⁾ with L₄ and L₃, pH = 8 for Cd(II)⁽²⁰¹⁾ with L₁, pH = 6 for Cd(II)⁽¹⁸⁵⁾ with L₄. The absorption of all chelate complexes solutions did not change over the whole range and very stable at pH = 5-8, The absorbance gradually decreased may be due to forming another chromophoric formula of reagent at higher pH⁽²⁰²⁾. The simplest explanation for these observations is probably that the ligand is highly protonated at low pH, making the nonbonding electron-pairs unavailable and therefore diminishing the tendency to form complexes with the metal cations. On the other hand, when the pH is above the optimum value, precipitation of copper(II), nickel(II) cobalt(II), chromium(III) and cadmium(II) ions become more significant and reduce the concentration of the complex⁽²⁰³⁾.

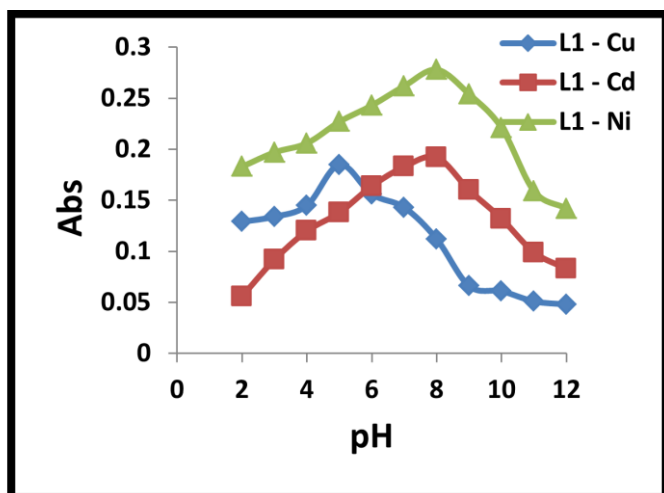


Figure (3.27): Effect of pH at $\lambda_{\text{max}} = 665 \text{ nm}$ for $L_1\text{-Cu}$, $\lambda_{\text{max}} = 650 \text{ nm}$ for $L_1\text{-Cd}$ and $\lambda_{\text{max}} = 540 \text{ nm}$ for $L_2\text{-Ni}$ complexes

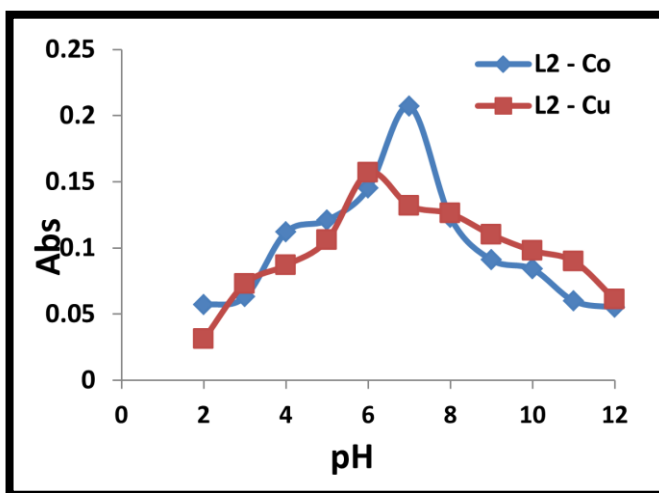


Figure (3.28): Effect of pH at $\lambda_{\text{max}} = 600 \text{ nm}$ for $L_2\text{-Co}$ and $\lambda_{\text{max}} = 647 \text{ nm}$ for $L_2\text{-Cu}$ complexes

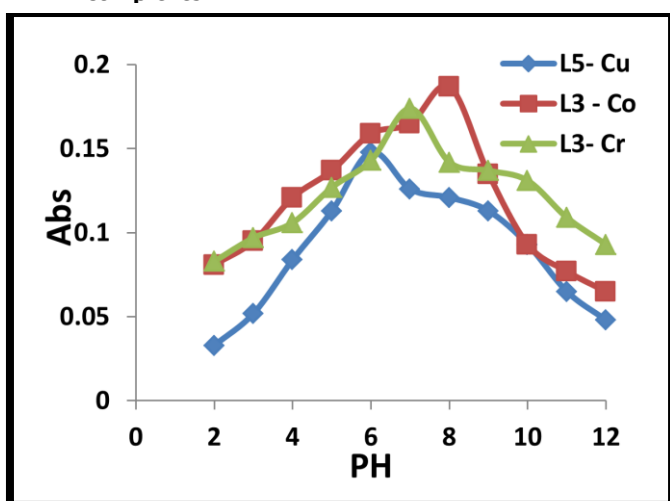


Figure (3.29): Effect of pH at $\lambda_{\text{max}} = 573 \text{ nm}$ for $L_3\text{-Cu}$, $\lambda_{\text{max}} = 566 \text{ nm}$ for $L_3\text{-Co}$ and $\lambda_{\text{max}} = 634 \text{ nm}$ for $L_3\text{-Cr}$ complexes

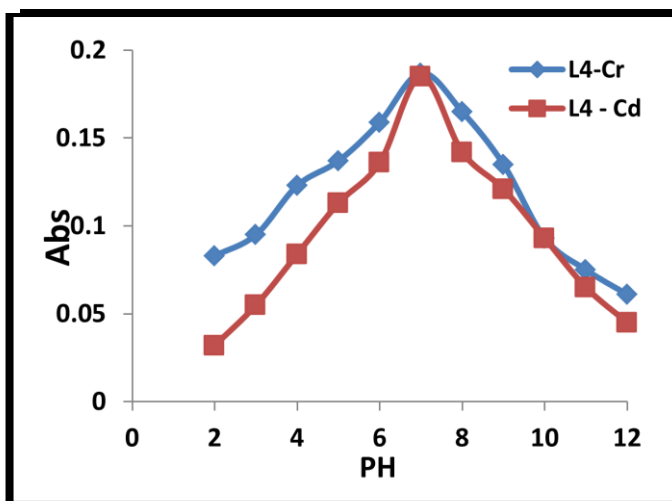


Figure (3.30): Effect of pH at $\lambda_{\text{max}} = 530 \text{ nm}$ for $L_4\text{-Cr}$ and $\lambda_{\text{max}} = 602 \text{ nm}$ for $L_4\text{-Cd}$ complexes

3.3: Determination of the stoichiometry of the complex

The continuous variation method (Job's method) was used for the determination of the stoichiometry of the complexes. A series of solutions were prepared, with constant molar concentration of metal and ligand. The absorbance recorded at λ_{max} . The results are showed that the M:L ratio for all complexes are 1:1, the results are tabulated in table (3.1) and figures (3.31) - (3.40) .

Table (3.1): Absorbance values at (λ_{max}) of complexes by continuous variation method (Job's method)

Seq.	V_L $1 \times 10^{-4} \text{ M}$	V_M $1 \times 10^{-4} \text{ M}$	V_M/V_{M+V_L}	$A_{L1- \text{Cu}}$	$A_{L1- \text{Cd}}$	$A_{L1- \text{Ni}}$	$A_{L2- \text{Co}}$	$A_{L2- \text{Cu}}$	$A_{L3- \text{Cu}}$	$A_{L3- \text{Cr}}$	$A_{L3- \text{Co}}$	$A_{L4- \text{Cd}}$	$A_{L4- \text{Cr}}$
1	1.8	0.2	0.10	0.051	0.073	0.063	0.054	0.053	0.080	0.085	0.092	0.063	0.093
2	1.6	0.4	0.20	0.064	0.116	0.094	0.065	0.087	0.124	0.112	0.121	0.082	0.120
3	1.4	0.6	0.30	0.078	0.143	0.132	0.114	0.126	0.153	0.136	0.147	0.128	0.146
4	1.2	0.8	0.40	0.108	0.167	0.153	0.173	0.158	0.189	0.158	0.185	0.148	0.167
5	1	1	0.50	0.125	0.192	0.197	0.213	0.185	0.223	0.175	0.223	0.188	0.193
6	1.2	0.8	0.60	0.112	0.165	0.148	0.175	0.147	0.175	0.161	0.173	0.164	0.173
7	1.4	0.6	0.70	0.082	0.134	0.124	0.136	0.120	0.148	0.138	0.159	0.132	0.152
8	1.6	0.4	0.80	0.061	0.098	0.092	0.091	0.082	0.119	0.117	0.126	0.112	0.148
9	1.8	0.2	0.90	0.049	0.081	0.077	0.069	0.045	0.076	0.090	0.093	0.087	0.135

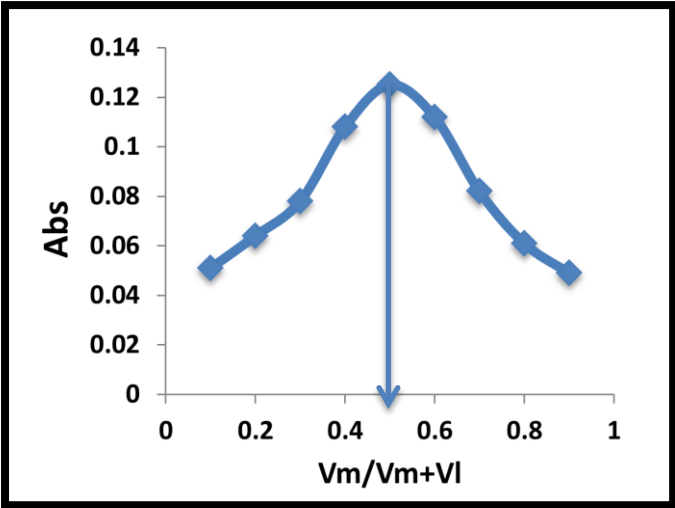


Figure (3.31) : Job’s method for the L₁-Cu at λ_{max} = 665nm

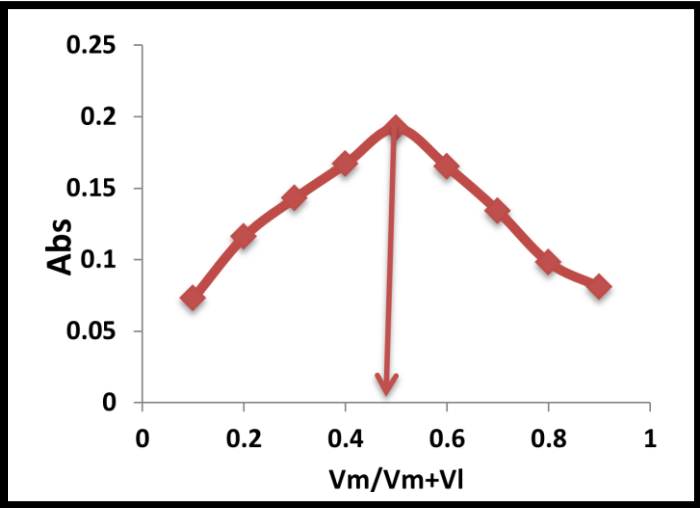


Figure (3.32) : Job’s method for the L₁-Cd at λ_{max} = 650nm

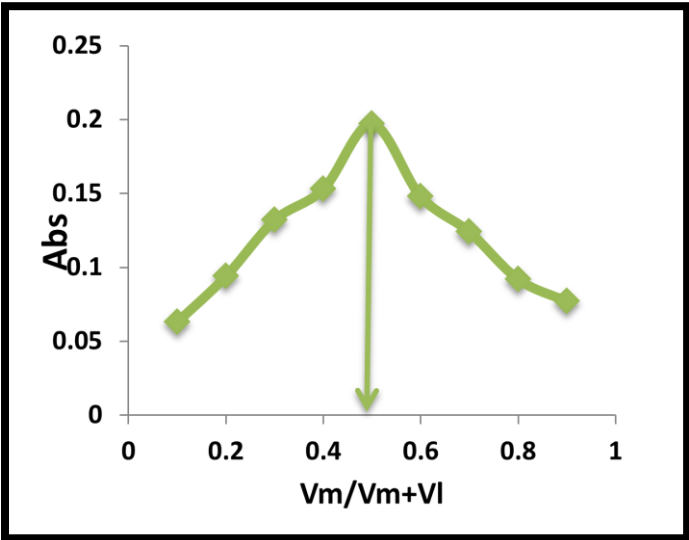


Figure (3.33) : Job’s method for the L₁-Ni at λ_{max}= 540 nm

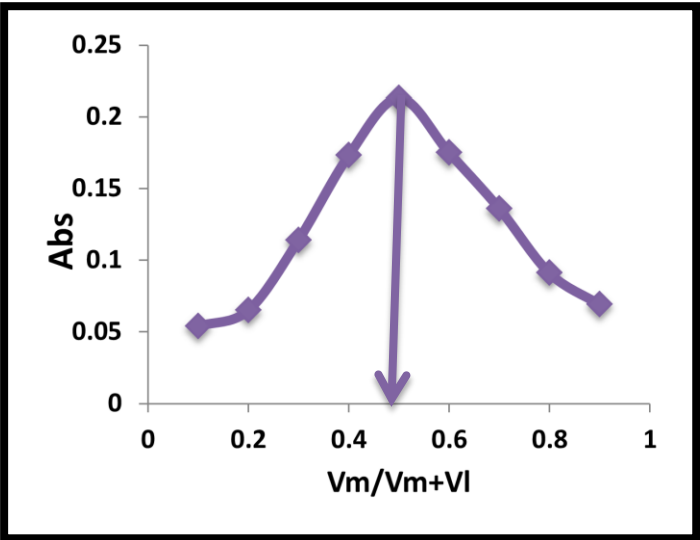


Figure (3.34) : Job’s method for the L₂-Co at λ_{max} = 600nm

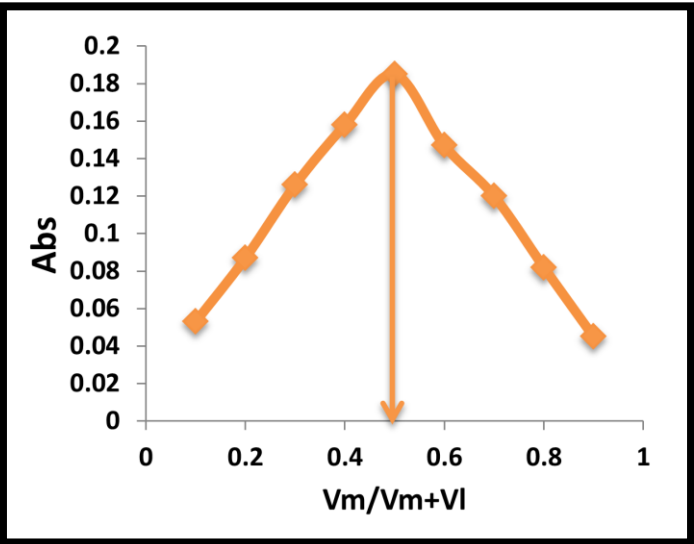


Figure (3.35) : Job’s method for the L₂-Cu at λ_{max}= 647nm

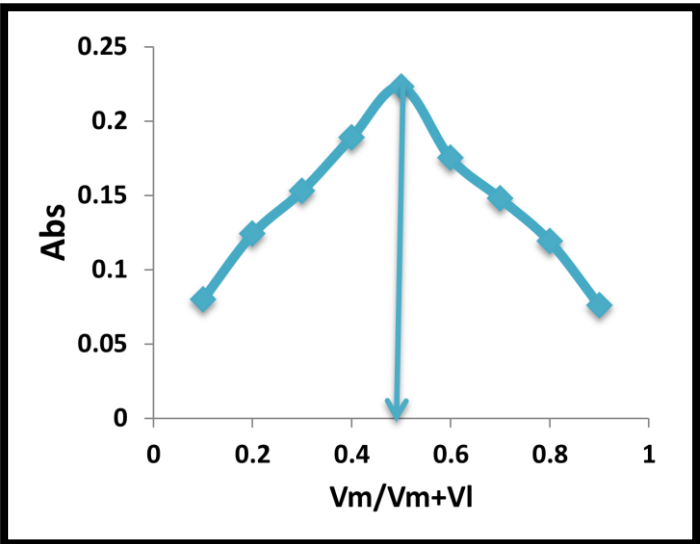


Figure (3.36) : Job’s method for the L₃-Cu at λ_{max} = 573nm

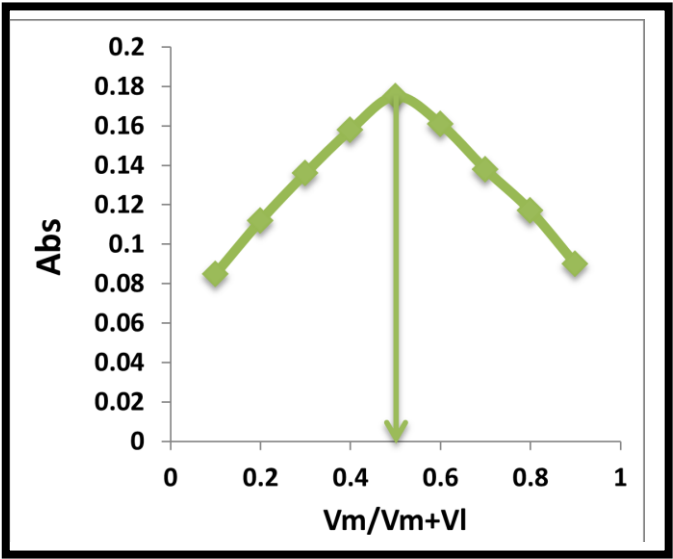


Figure (3.37): Job's method for the L₃-Cr at λ_{max} = 634nm

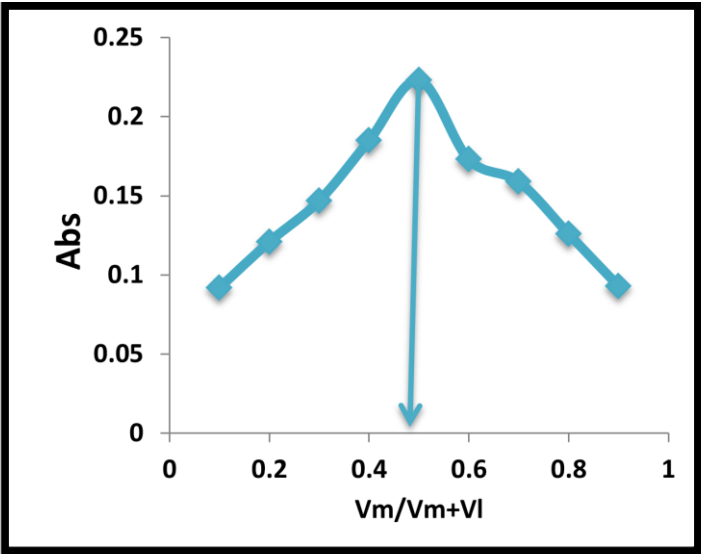


Figure (3.38): Job's method for the L₃-Co at λ_{max} =566 nm

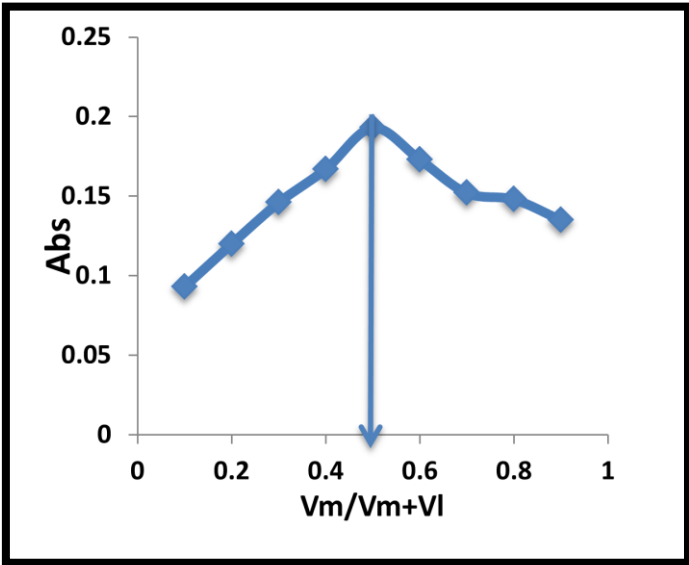


Figure (3.39): Job's method for the L₄-Cr at λ_{max} =530 nm

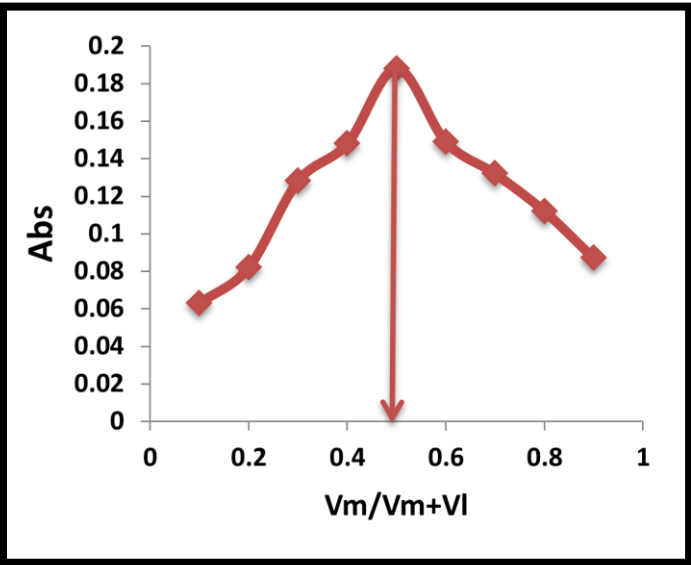


Figure (3.40): Job's method for the L₄-Cd at λ_{max} = 602 nm

3.4: Calculation of the Metal Complexes Stability Constant (K_{st})

Stability constant of complexes were obtained spectrophotometrically by measuring the absorbance of solutions of reagent and metal ion at fixed wavelength λ_{max} , and fixed pH. The degree of formation of the complexes(K_{st}) were obtained from the relationship of $K_{st} = (1-\alpha / \alpha^2C)$ for 1:1 (M:L) metal complex and $\alpha = A_m - A_s / A_m$, where A_m and A_s are the absorbance of equivalent amount of L:M and excess amount of ligand with constant volume of metal formed complex respectively⁽¹⁸⁴⁾. The calculated K_{st} values for the complexes are given in table (3.2). The values of stability constant indicated high stability complexes⁽²⁰⁰⁾.

Table (3.2): Stability Constant Values (K_{st}) at (λ_{max}) and pH of Suggested Complexes

Complexes	λ_{max} (nm)	pH	A_s	A_m	α	K_{st}
[CuL ₁ Cl ₂]	665	5	0.158	0.183	0.137	0.459×10^6
[CdL ₁ Cl ₂]	650	8	0.167	0.222	0.248	0.122×10^6
[NiL ₁ Cl ₂]	540	8	0.112	0.138	0.188	0.230×10^6
[CoL ₂ Cl ₂]	600	7	0.182	0.232	0.216	0.167×10^6
[CuL ₂ Cl ₂]	647	6	0.176	0.210	0.162	0.320×10^6
[CrL ₃ Cl ₂]Cl	634	7	0.145	0.211	0.313	0.701×10^5
[CuL ₃ Cl ₂]	573	6	0.133	0.162	0.179	0.257×10^6
[CoL ₃ Cl ₂]	566	8	0.144	0.188	0.234	0.139×10^6
[CdL ₄ Cl ₂]	602	6	0.082	0.108	0.241	0.131×10^6
[CrL ₄ Cl ₂]Cl	530	7	0.065	0.088	0.261	0.109×10^6

3.5: Infrared spectra of the ligands and their suggested complexes

The IR spectrum was obtained for azo-azomethine ligands prepared as KBr disk but the complexes as CsI disk by using a Shimaduz FT-IR spectrophotometer. Figures (3.41) - (3.48) and table (3.3) summarize the IR data of the synthesized ligands and their complexes. Similar prepared azo-azomethine ligands (L_1 - L_4) to the active groups, which are clearly shown in infrared spectra. These compounds contain groups (C=N), (N=N) and (OH), in all these compounds absorbance bands in the spectral range $(1627-1639) \text{ cm}^{-1}$ due to the stretching vibration of the (C=N)^(183,194) also owns bands in the range $(1465-1481) \text{ cm}^{-1}$ due to the stretching vibration of the (N=N)⁽²⁰²⁾, while the stretching vibration band of the (O-H)⁽⁴⁶⁾ group appeared broad in the range $(3208 - 3336) \text{ cm}^{-1}$, the other hand azo-azomethine ligands have strong bands within the range $(1543-1590) \text{ cm}^{-1}$ due to stretching vibration of the bond (C=C) of the aromatic rings⁽²⁰⁴⁾. As well as all prepared ligands containing the bands in the range $(3056 - 3093) \text{ cm}^{-1}$ due to the stretching vibration for a group (C-H aromatic)^(204,205), and contains a band at $(2931 - 2970) \text{ cm}^{-1}$ due to the stretching vibration for a group (C-H aliphatic)⁽²⁰⁵⁾. In ligand L_1 contains other bands which appear at $(1133 \text{ and } 833) \text{ cm}^{-1}$ due to stretching vibration for the group (C-S)⁽¹⁹¹⁾ and a strong band in the position 1600 cm^{-1} due to stretching vibration for the (C=N)⁽²⁰⁶⁾ in thiozole ring.

Infrared spectra are very important in determent the type of correlation in the case of complexes, where appeared in the complexes stretching vibration band for the groups (OH) are broad in the range $(3217-3405)^{(185)} \text{ cm}^{-1}$, this indicates the association of this group with the central metal atom, where the correlation of the oxygen atom with the metal atom is an M-O component. The stretching vibration band in the range $(1612-1626) \text{ cm}^{-1}$ due to (C=N) group in the complexes. The shift of this band to a lower wave numbers compared to its position in the ligand

can be referred to a shift of lone pair density toward the metal ion, such shift supports the participation of the azomethine nitrogen in coordination⁽²⁰⁵⁾.

Complexes spectrum identify appearance of new bands which were not present in the spectrum of ligand due to stretching vibration (M-N), (M-O) and (M-Cl), where appeared band (M-O)^(206,207) in the range (476 - 578) cm^{-1} , while stretching vibration band of the (M-N)^(206,207) appeared at (416 - 486) cm^{-1} and appeared band in the range (331–378) cm^{-1} due to stretching vibration for the (M-Cl). In addition, the vibration band of the azo chromogen group both in the ligands and the complexes were observed at (1473,1481) cm^{-1} respectively. Although the azo chromogen group has a donor character and can form coordination compounds, the constant value of its vibration both in the ligand and the complexes shows that there is no coordination between the azo chromogen group and the metal ions⁽²⁰⁷⁾. All the vibrational data proposes that the metal ions are bonded to the azo-azomethine ligand through the phenolic oxygen and the imino nitrogen atoms⁽²⁰⁵⁾.

Table (3.3): FT-IR spectra of the azo-azomethine and complexes

Comp.	C=N azom. C=N thiaz.	N=N	C=C	OH	C-H arom.	C-H aliph.	M-O	M-N	M-Cl
L₁	1627 s. 1600 s.	1477s.	1554s.	3336 br.	3056 w.	2939 w.	-	-	-
L₂	1630 s. -	1481s.	1543m.	3294 br.	3093 w.	-	-	-	-
[CoL₂Cl₂]	1626 s. -	1481s.	1535	3281 br.	3078 w.	-	547m.	462 m.	378 m.
[CuL₂Cl₂]	1612 s. -	1481v.s. .	1533m.	3276 br.	3062 w.	-	476m.	416 m.	331 s.
L₃	1639 v.s. -	1465v.s. .	1582v.s. .	3208 br.	3059 w.	2970 w.	-	-	-

L_4	1633 v.s. -	1473v.s .	1590v.s .	3306 br.	3067 w.	2931 w.	-	-	-
$[CrL_4Cl_2]$	1619 v.s. -	1473s.	1556s.	3217w. - 3405br.	3065 w.	2966 w.	532- 578 m.	432- 486s .	308- 362s.
$[CdL_4Cl_2]$	1622 s. -	1473s.	1555m.	3340- 3402br.	3066 w.	2962 w.	563m.	424- 486s .	331- 383s.

br. = broad, v.s = very strong, s = strong, m = medium, w= weak

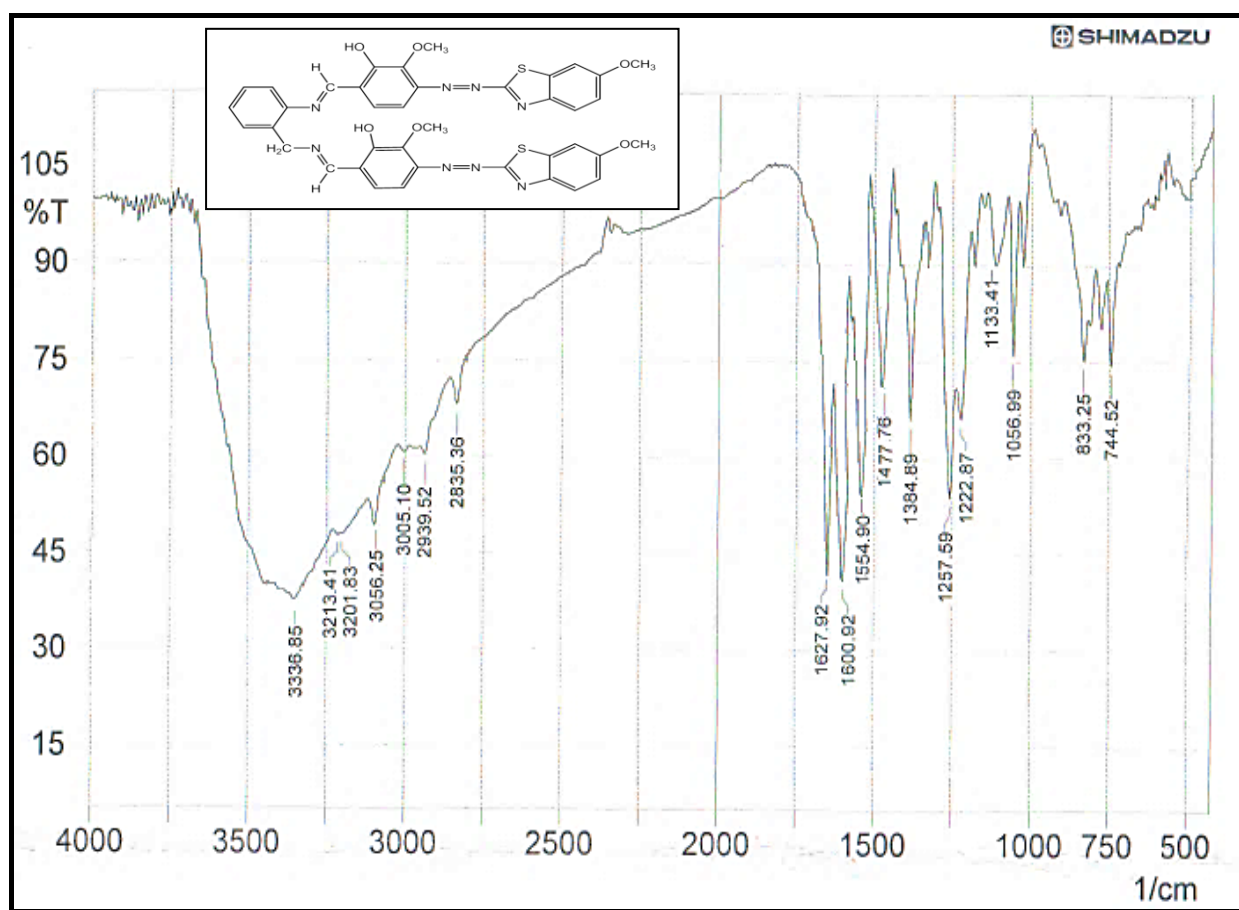
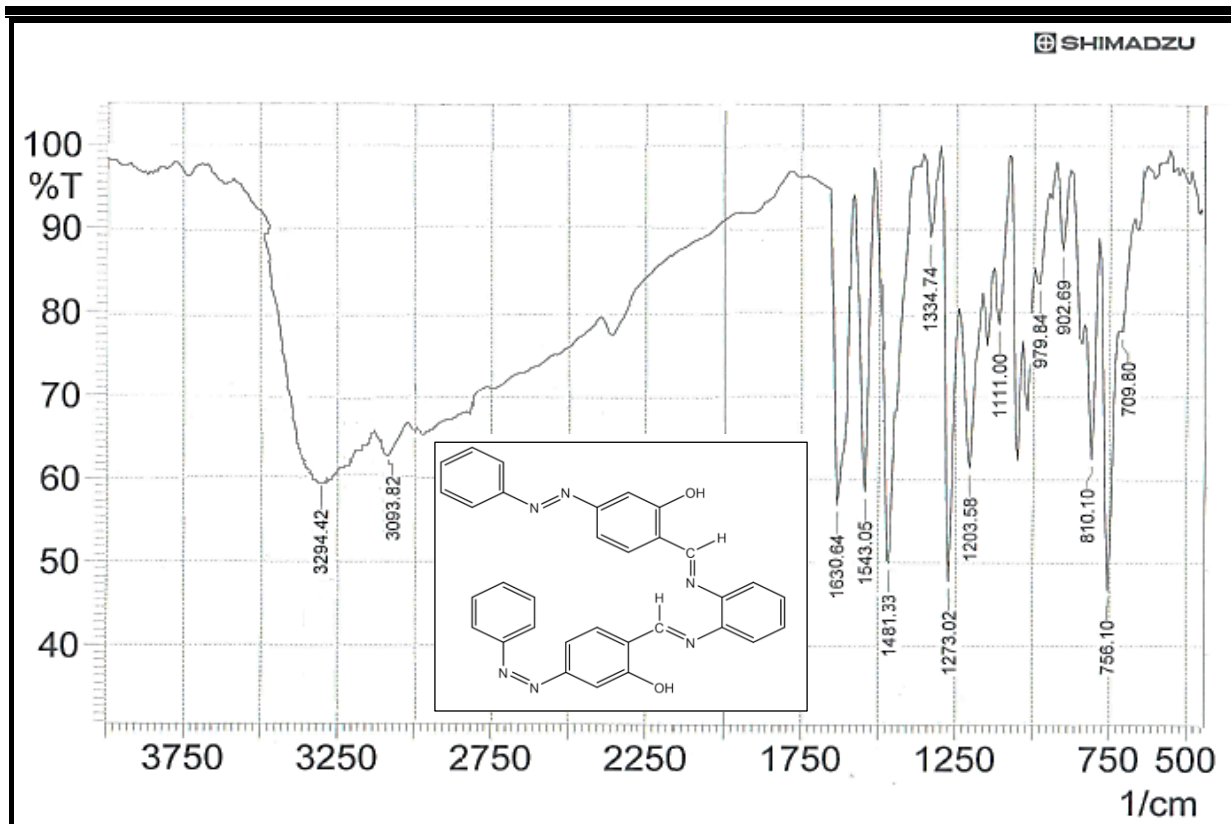
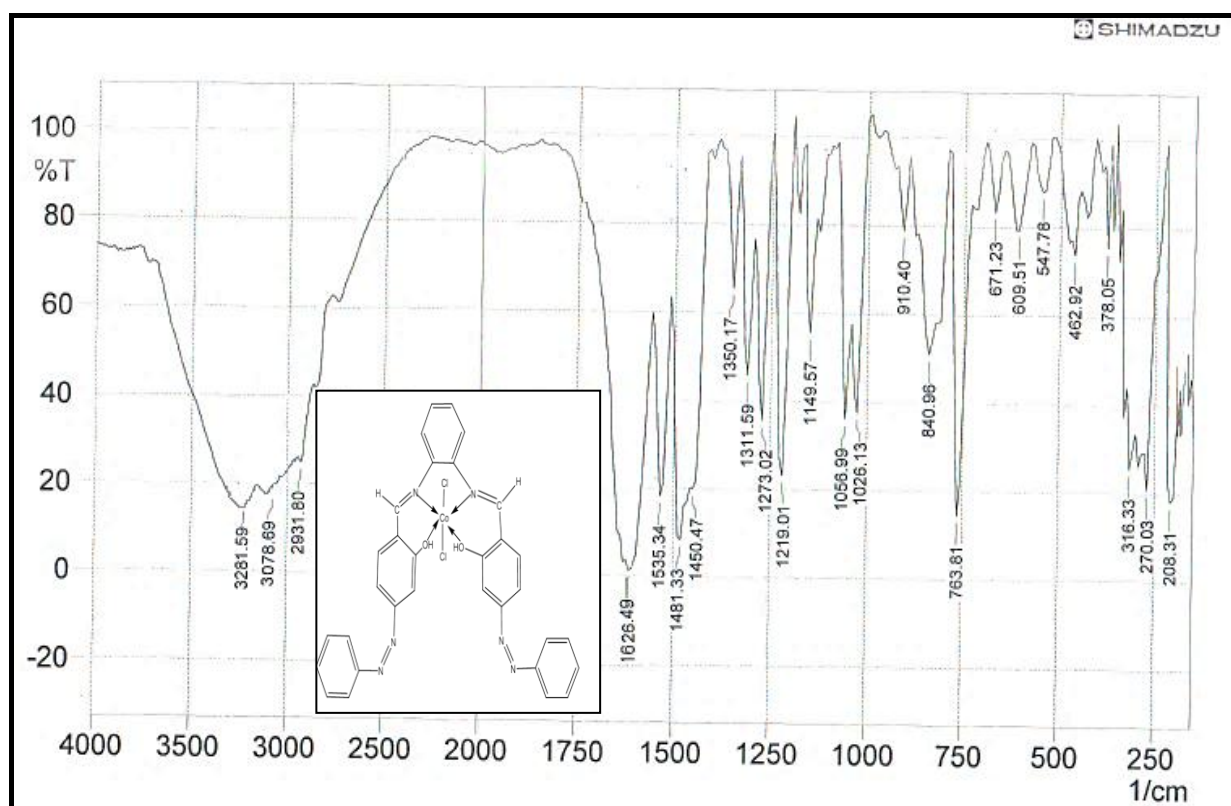
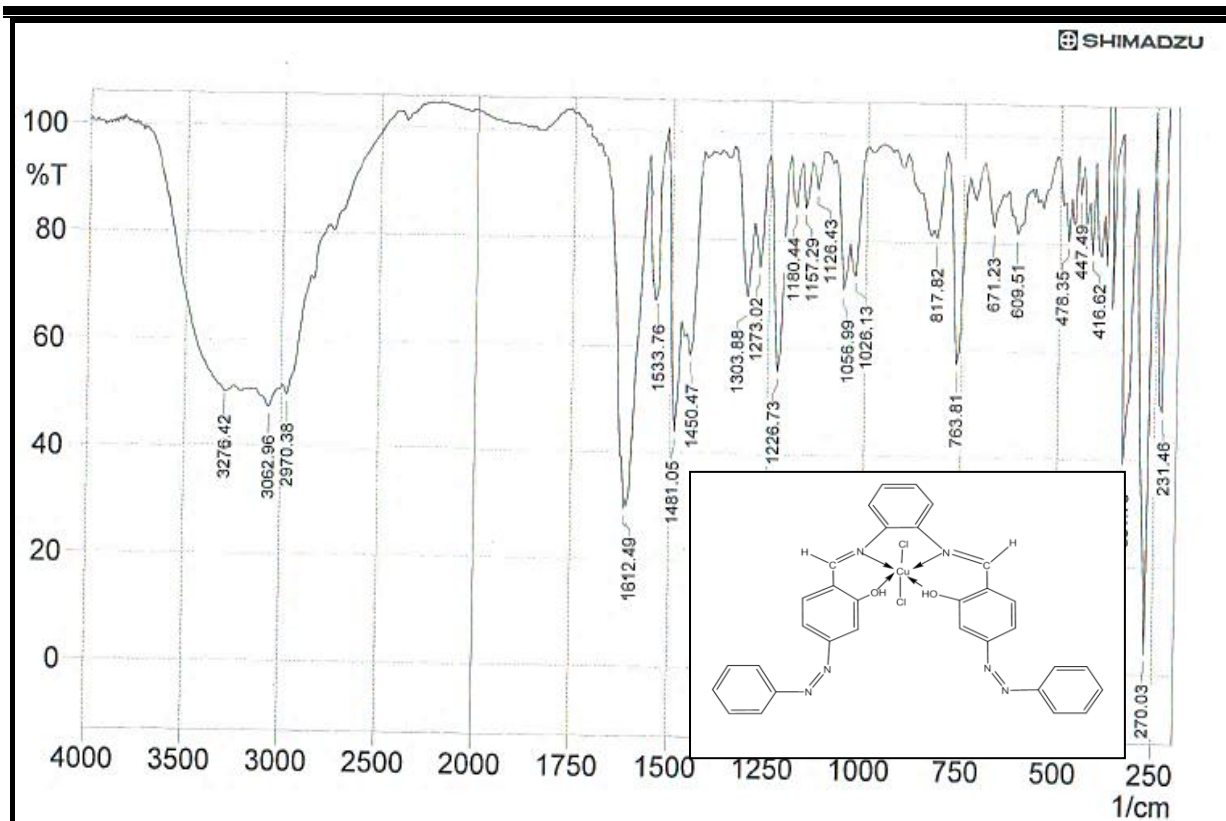
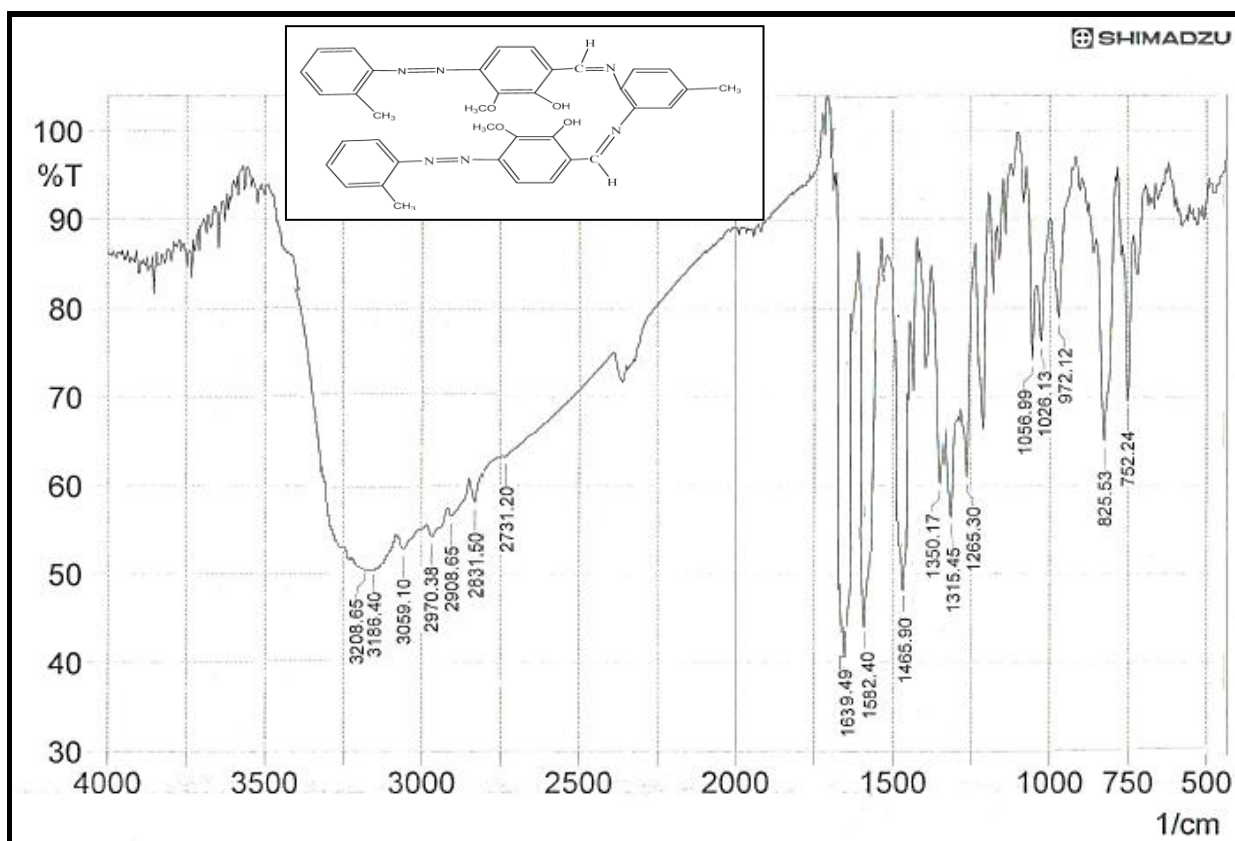
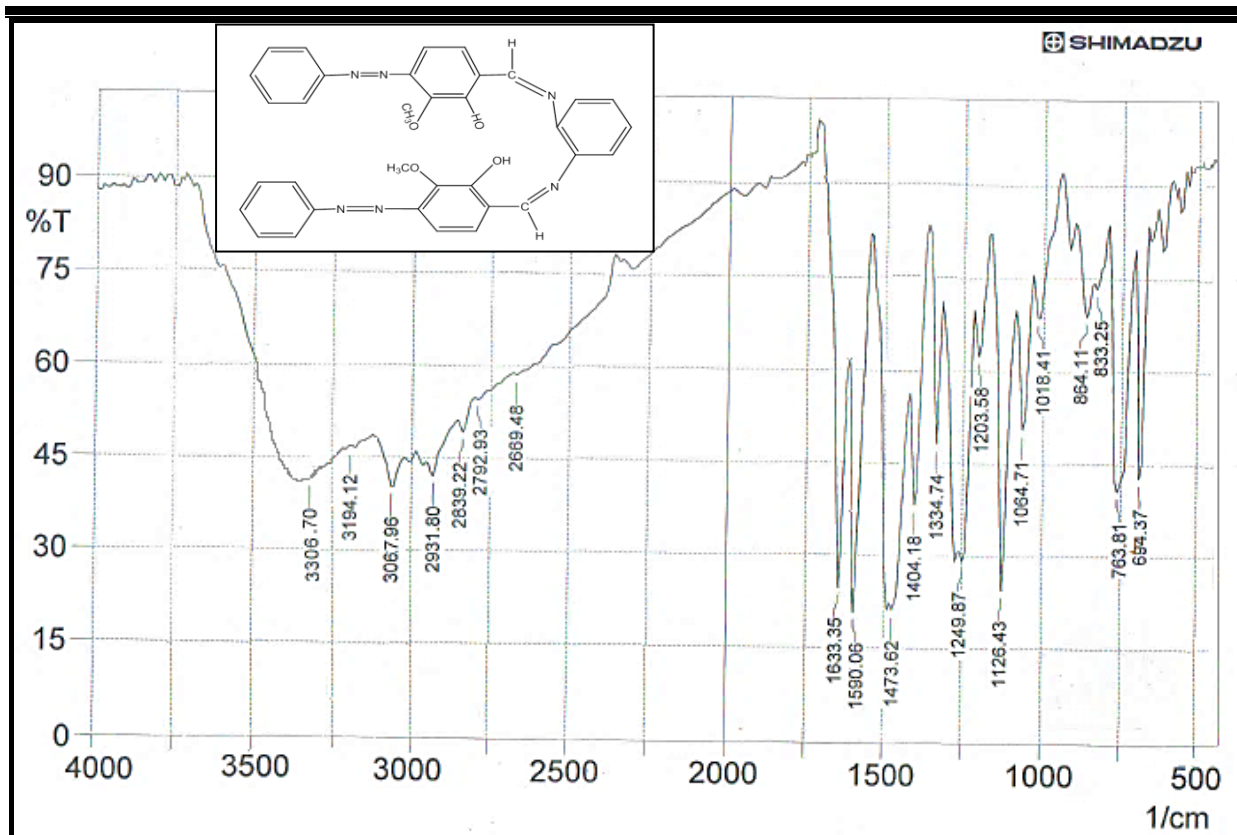
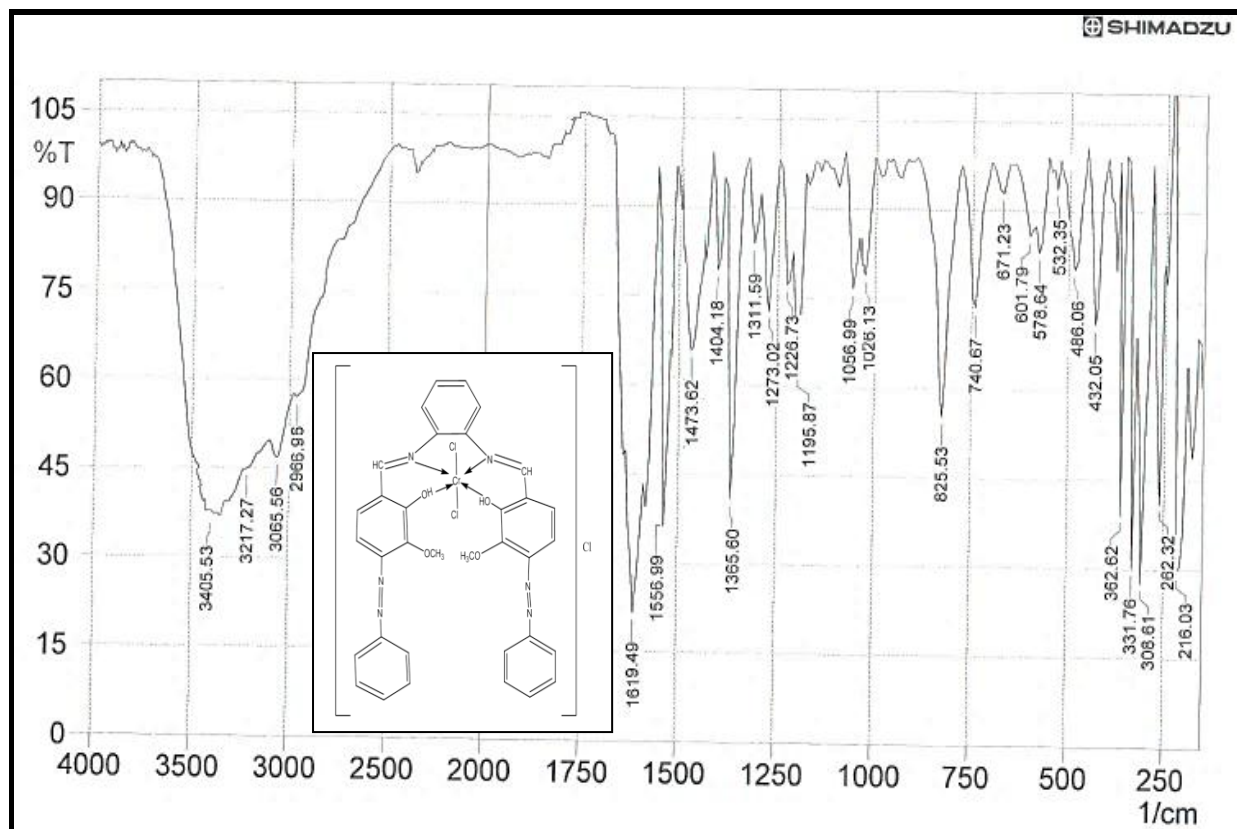


Figure (3.41): IR spectrum of L_1 by KBr

Figure (3.42): IR spectrum of L_2 by KBrFigure (3.43): IR spectrum of $[\text{Co}L_2\text{Cl}_2]$ by CsI

Figure (3.44): IR spectrum of $[\text{CuL}_2\text{Cl}_2]$ by CsIFigure (3.45): IR spectrum of L_3 by KBr

Figure (3.46): IR spectrum of L_4 by KBrFigure (3.47): IR spectrum of $[\text{Cr}L_4\text{Cl}_2]\text{Cl}$ by CsI

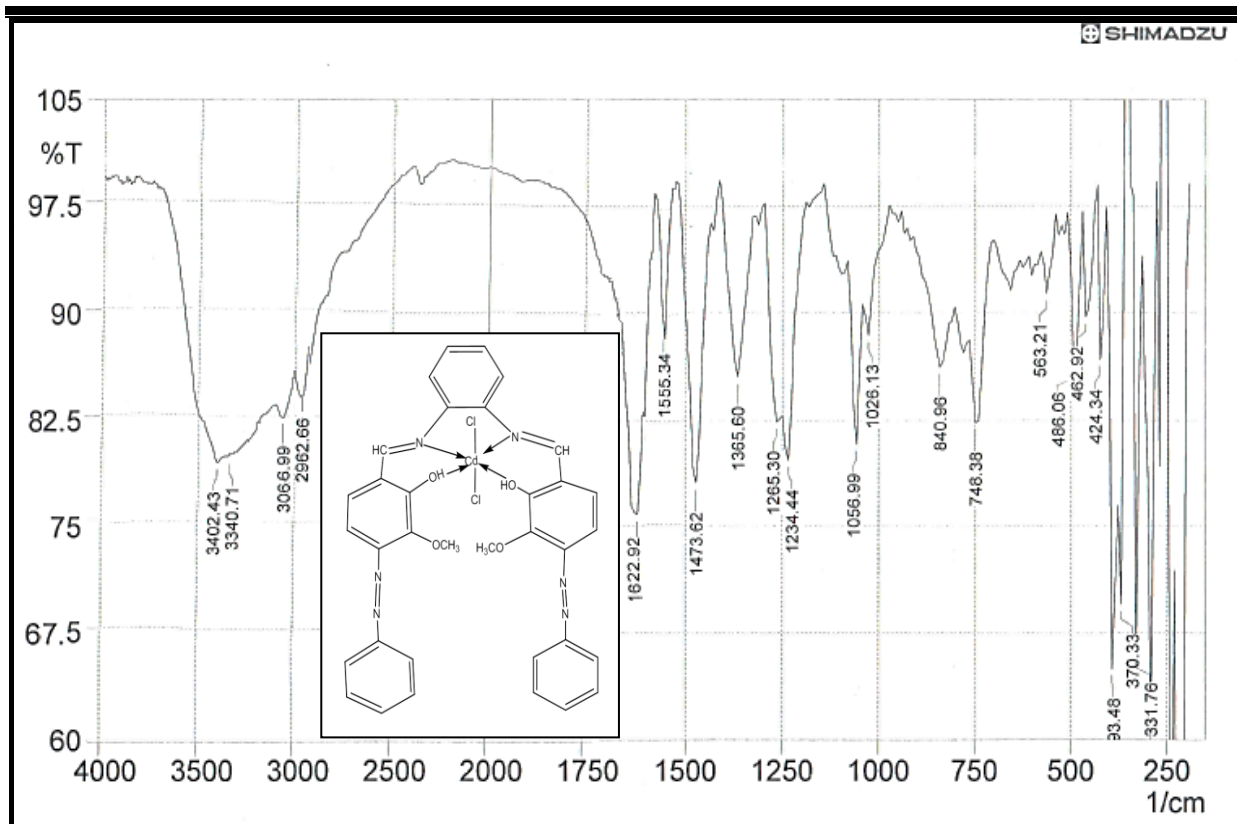


Figure (3.48): IR spectrum of $[\text{CdL}_4\text{Cl}_2]$ by CsI

3.6: ^1H -NMR spectrum

The ^1H -NMR spectrum of the prepared azo-azomethine compounds (L_1 - L_4) is shown in figures (3.49) - (3.52). The ^1H -NMR spectrum appearance of multiplet signal at chemical shift (δ) (6.7-7.9) ppm due to aromatic ring protons (Ar-H)⁽²⁰⁸⁾, the appearance of a singlet signal at 4.9 ppm due to (CH_2) ⁽²⁰⁹⁾ in L_1 , while azomethine proton⁽²⁰⁸⁾ was appeared in rang of (7.8-8.1) ppm and the appearance a singlet signal at (at 8.3-10.1)ppm due to $(-\text{OH})$ ⁽⁴⁶⁾. The azo-azomethine compounds (L_1, L_3 and L_4) characterized by the appearance of a singlet signal at (3.7 - 4.0) ppm due to $(-\text{OCH}_3)$ ^(208,210) in L_1, L_3 and L_4 and show the a singlet signal in the L_3 at (3.8) ppm due to $(-\text{CH}_3)$ ⁽²¹¹⁾.

Table (3.4): ¹H-NMR spectra data of prepared azo-azomethine compounds

Symbol	δ(ppm)					
	-OCH ₃	-CH ₂	-CH(aromatic)	-CH ₃	-CH=N	Ar-OH
L ₁	3.7, 3.8	4.9	6.7-7.6	-	7.8	8.6-8.9
L ₂	-	-	7.5-7.9	-	8.1	10.1
L ₃	4.0	-	6.9-7.7	3.8	7.9	8.3
L ₄	3.9	-	7.2-7.6	-	7.8	8.4

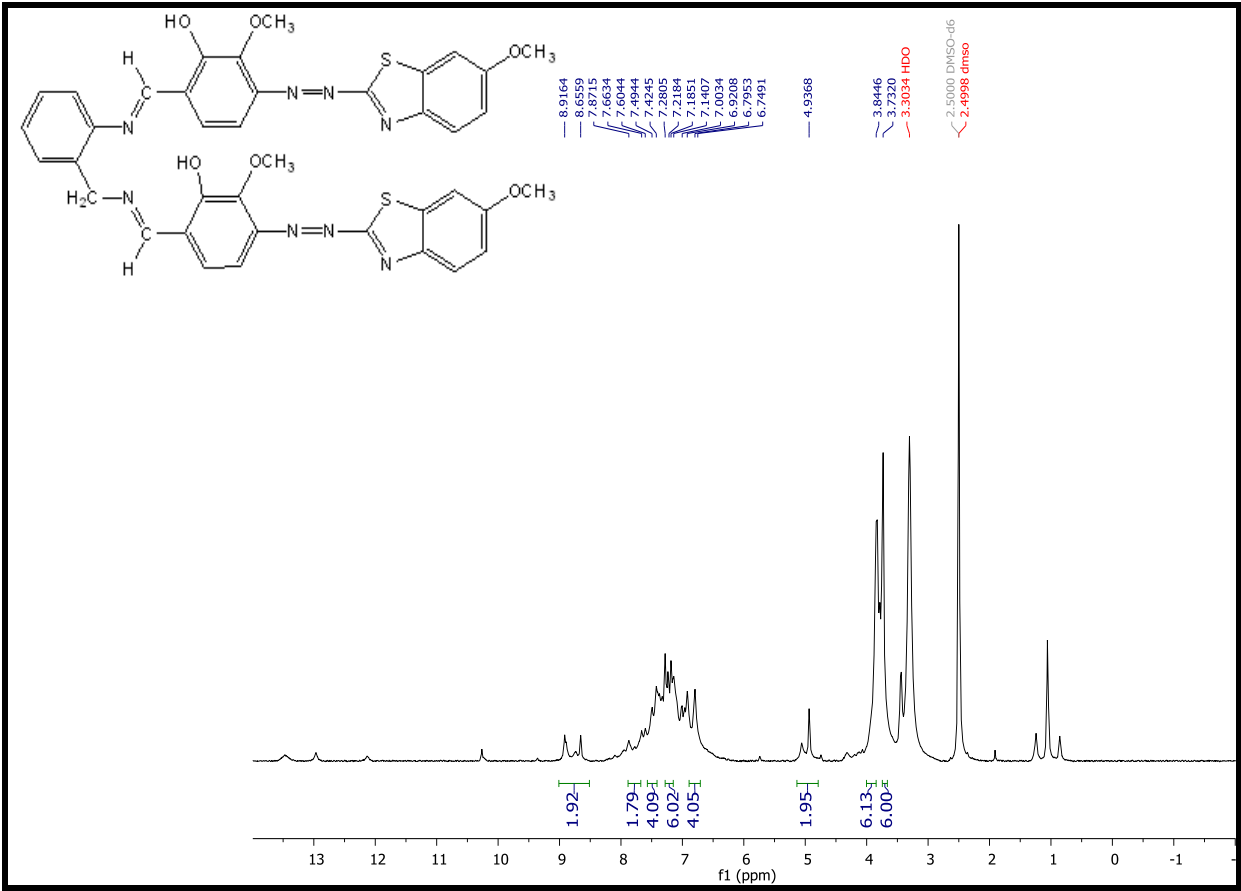


Figure (3.49): ¹H-NMR spectrum of the L₁

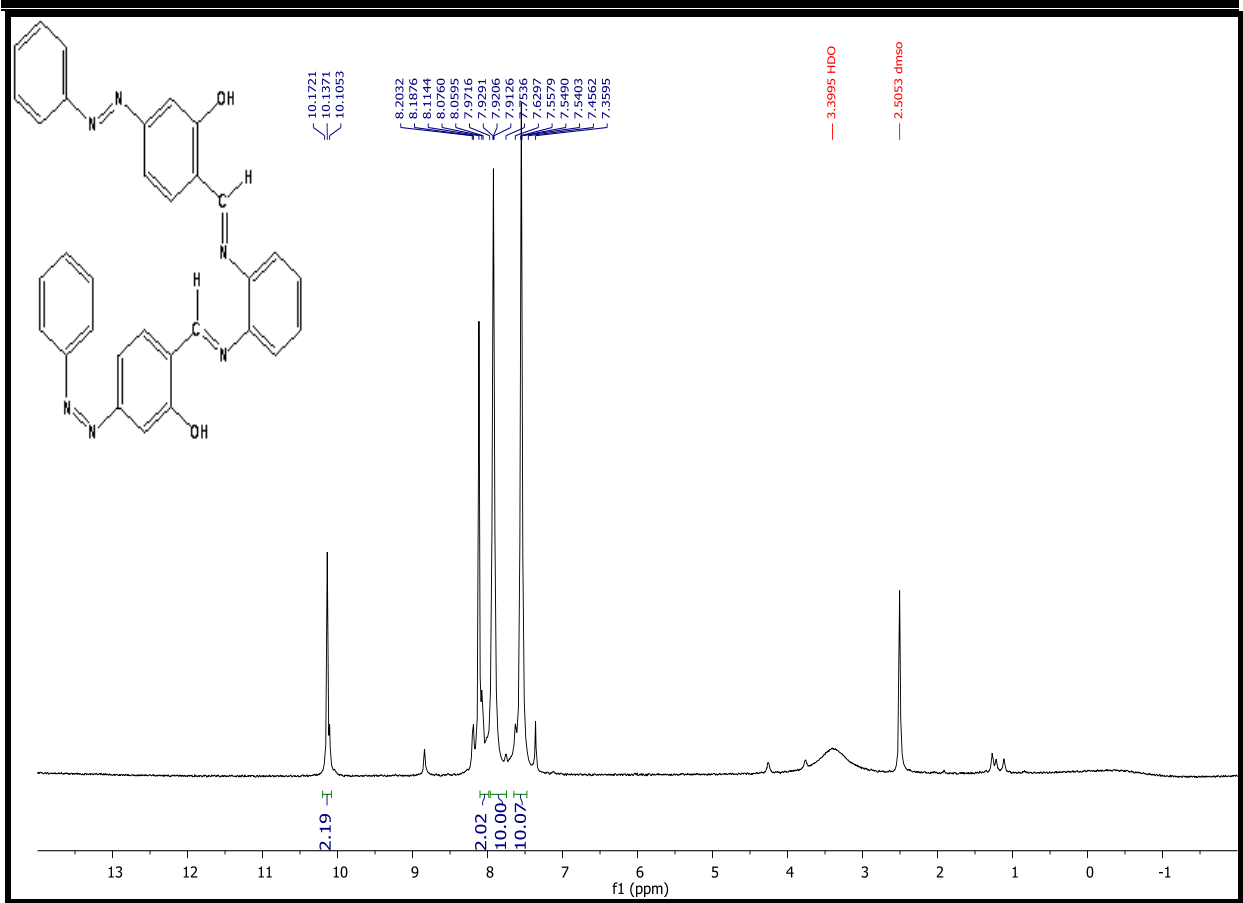


Figure (3.50): ¹H-NMR spectrum of the L₂

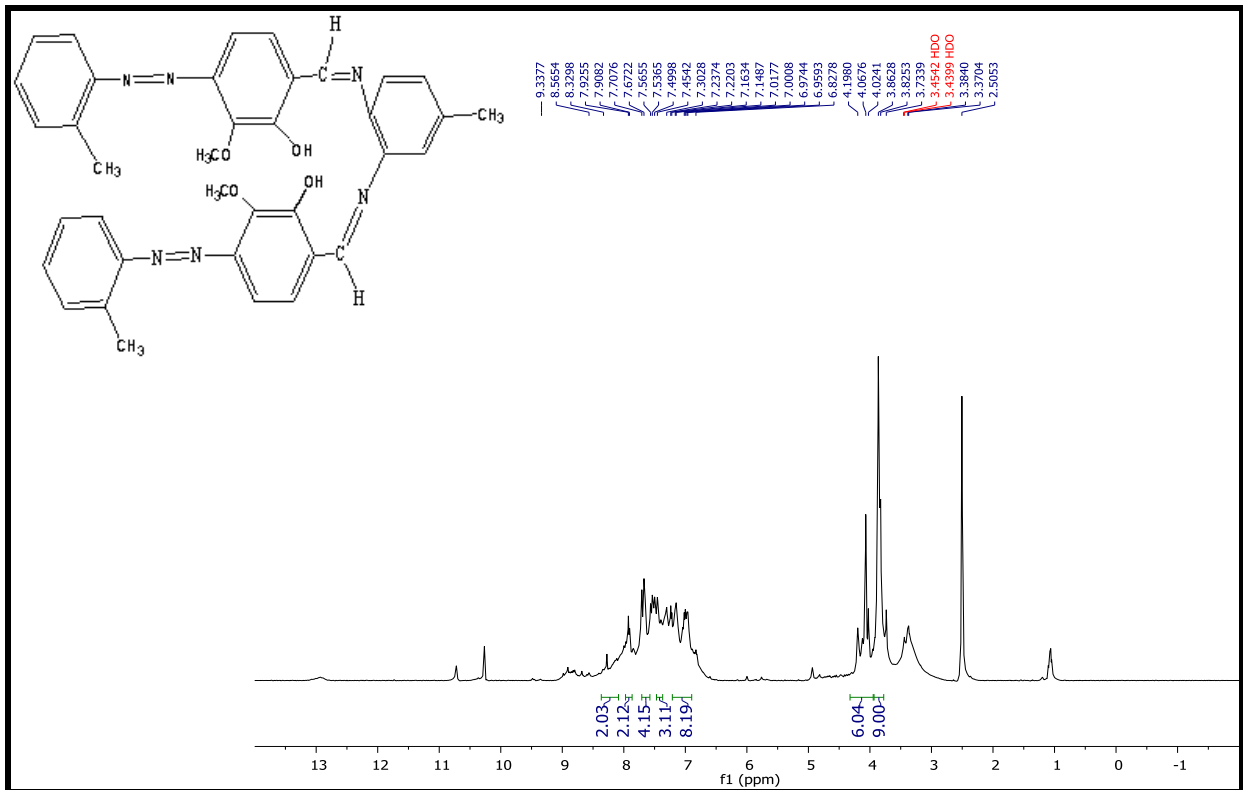


Figure (3.51): ¹H-NMR spectrum of the L₃

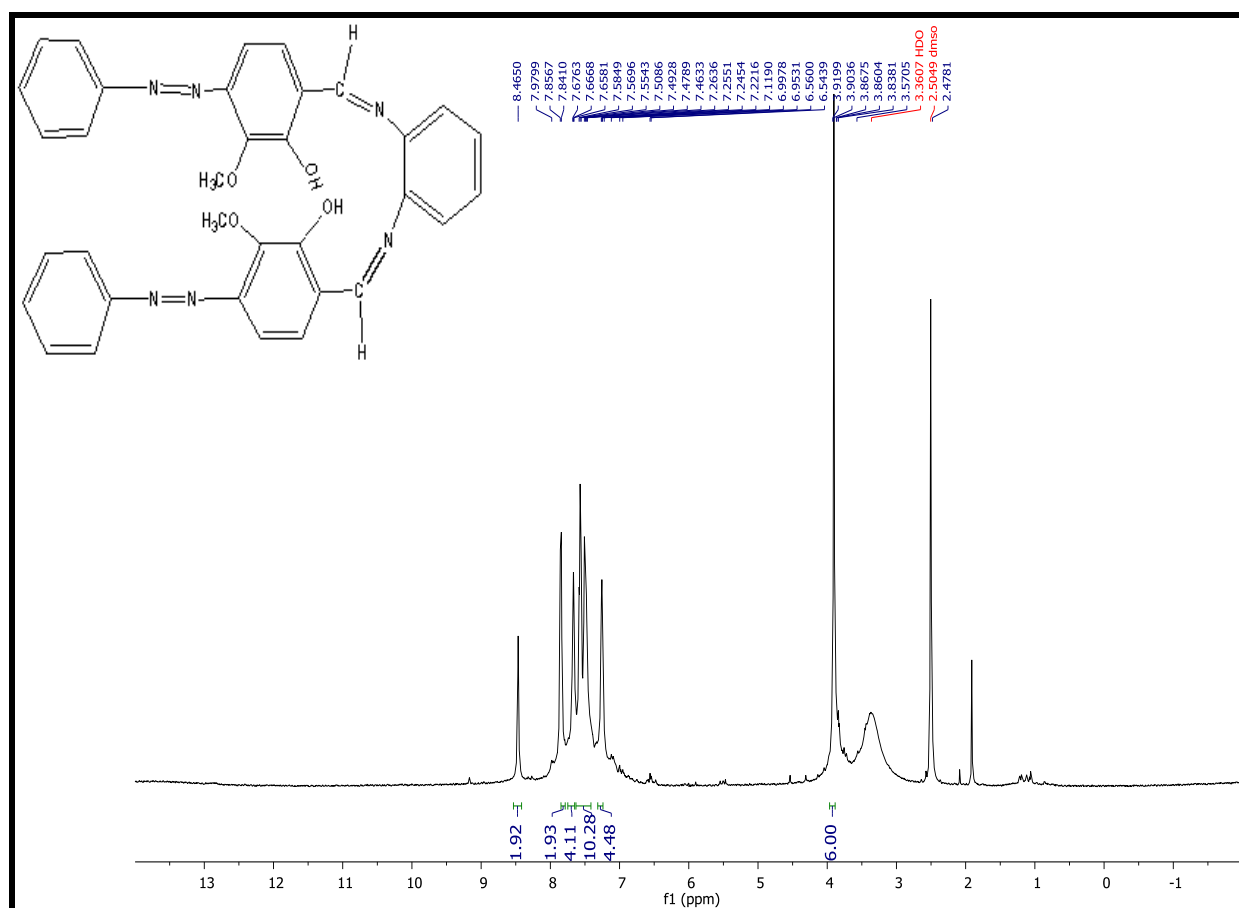


Figure (3.52): ^1H -NMR spectrum of the L_4

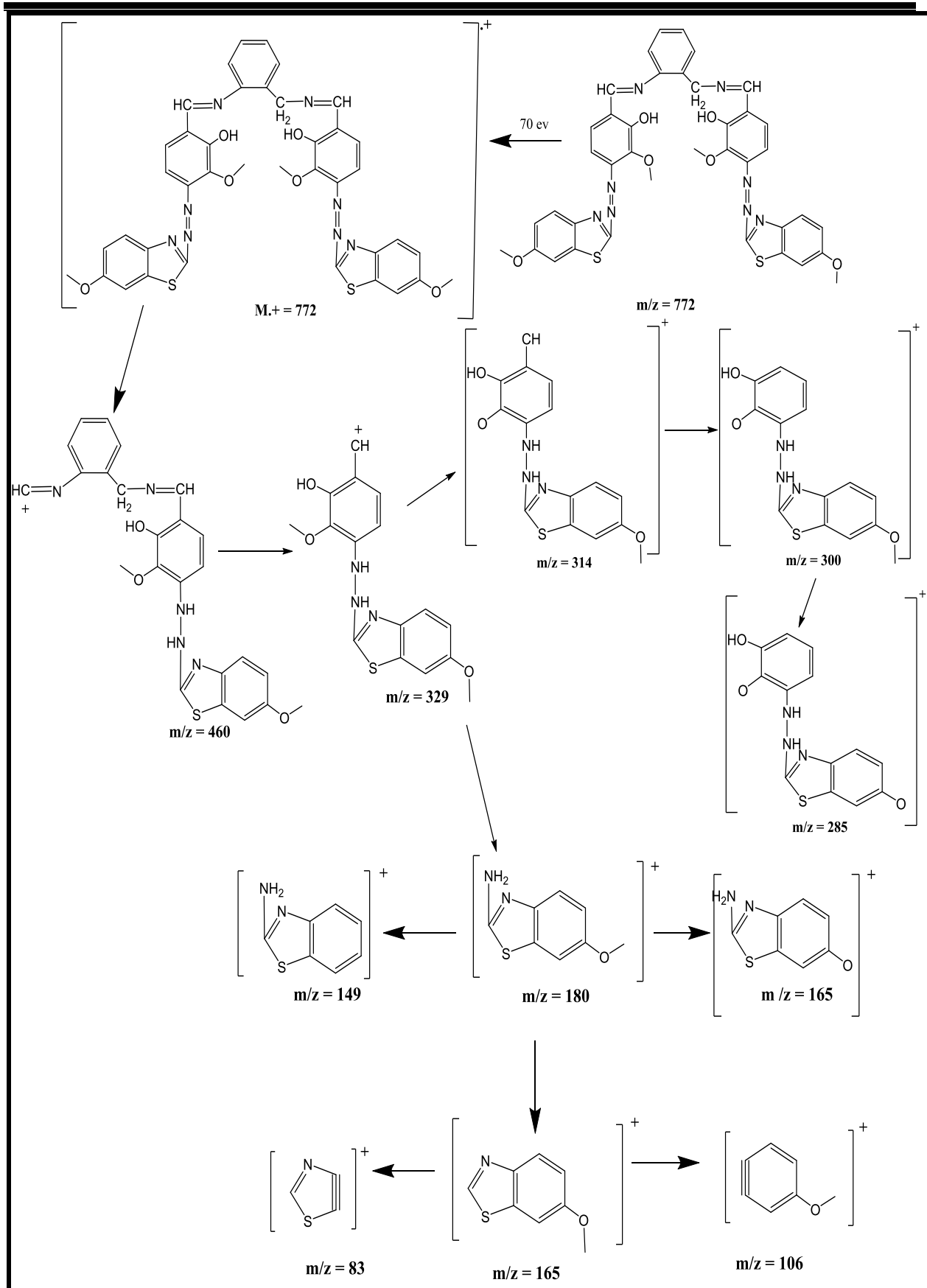
3.7: Mass spectra of Azo - Azomethine

3.7.1: Mass spectra of L_1

Characterize mass spectra of the ligand appearance of molecular ion peak (M^+) at 772 m/z and the spectrum show a series of peaks (figure 3.53) at m/z = 460, 329, 314, 300, 285, 180, 165, 149, 106 and 83 corresponding to $[\text{C}_{24}\text{H}_{22}\text{N}_5\text{O}_3\text{S}]^+$, $[\text{C}_{16}\text{H}_{15}\text{N}_3\text{O}_3\text{S}]^+$, $[\text{C}_{15}\text{H}_{12}\text{N}_3\text{O}_3\text{S}]^+$, $[\text{C}_{14}\text{H}_9\text{N}_3\text{O}_3\text{S}]^+$, $[\text{C}_{13}\text{H}_7\text{N}_3\text{O}_3\text{S}]^+$, $[\text{C}_8\text{H}_8\text{N}_2\text{OS}]^+$, $[\text{C}_7\text{H}_5\text{N}_2\text{OS}]^+$, $[\text{C}_7\text{H}_5\text{N}_2\text{S}]^+$, $[\text{C}_7\text{H}_6\text{O}]^+$ and $[\text{C}_3\text{NHS}]^+$ respectively. list in table (3.5). The mechanics of break- up are embedded in the scheme (3.1).

Table (3.5): Includes the important peaks which attributed to the fragment of L₁

m/z	
772	[C ₃₉ H ₃₂ N ₈ O ₆ S ₂]
460	[C ₂₄ H ₂₂ N ₅ O ₃ S] ⁺
329	[C ₁₆ H ₁₅ N ₃ O ₃ S] ⁺
314	[C ₁₅ H ₁₂ N ₃ O ₃ S] ⁺
300	[C ₁₄ H ₉ N ₃ O ₃ S] ⁺
285	[C ₁₃ H ₇ N ₃ O ₃ S] ⁺
180	[C ₈ H ₈ N ₂ OS] ⁺
165	[C ₇ H ₅ N ₂ OS] ⁺
149	[C ₇ H ₅ N ₂ S] ⁺
106	[C ₇ H ₆ O] ⁺
83	[C ₃ NHS] ⁺

Scheme (3.1): Mass spectra for the ligand L_1

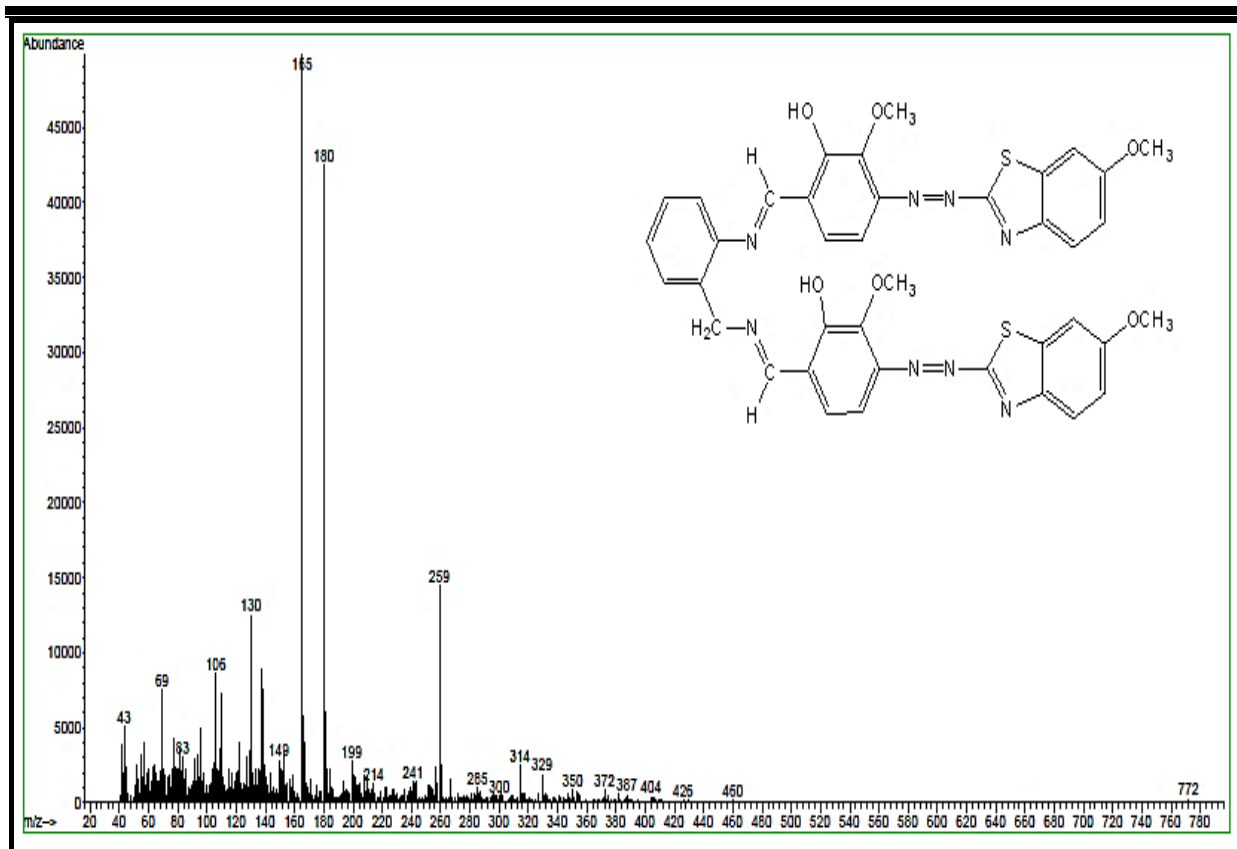


Figure (3.53): Mass spectrum of the L_1

3.7.2 : Mass spectra of L_2 and their complexes

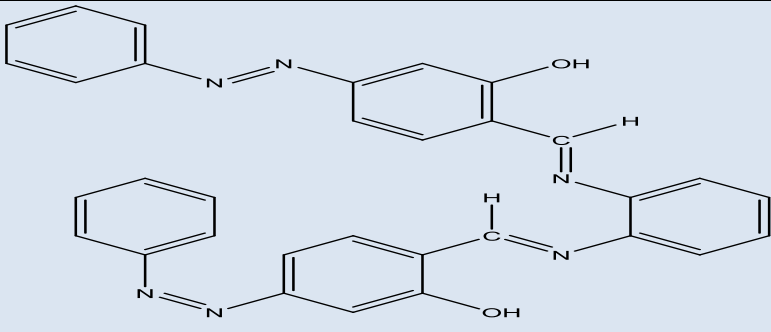
Characterize mass spectra of the ligand appearance of molecular ion peak (M^+) at 523 m/z and the spectrum show a series of peaks (figure 3-54) at m/z = 432, 419, 314, 298, 197, 182, 133, 103, 91 and 76 corresponding to $[C_{26}H_{19}N_5O_2]^+$, $[C_{26}H_{19}N_4O_2]^+$, $[C_{20}H_{14}N_2O_2]^+$, $[C_{20}H_{13}N_2O]^+$, $[C_{12}H_9N_2O]^+$, $[C_{12}H_{10}N_2]^+$, $[C_7H_5N_2O]^+$, $[C_7H_5N]^+$, $[C_6H_5N]^+$ and $[C_6H_4]^+$ respectively. list in table (3.6). The mechanics of break- up are embedded in the scheme (3.2).

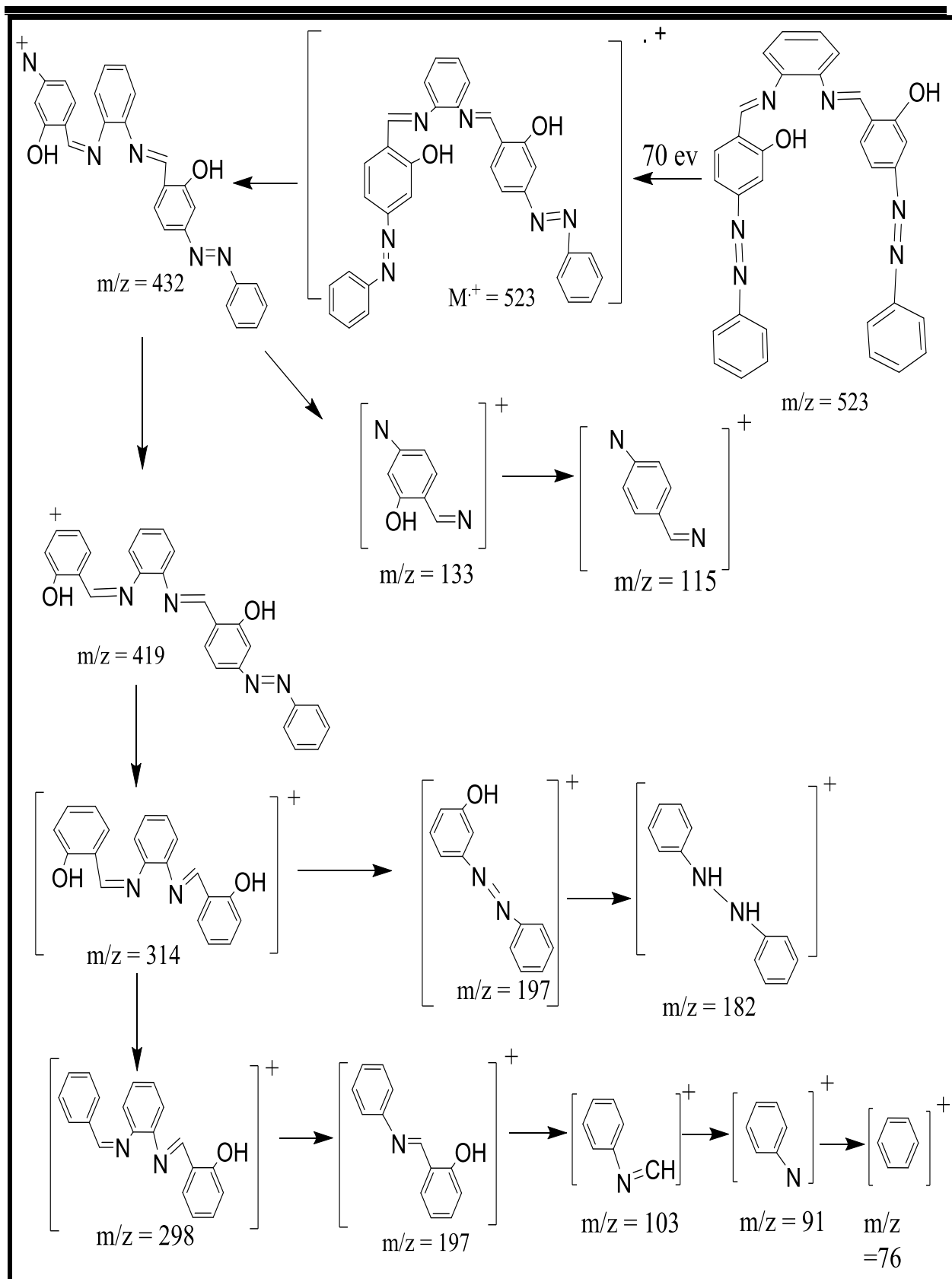
The mass spectra of the complex $[CuL_2Cl_2]^+$ (figure 3.55) shows of molecular ion (M^+) at 659 m/z and appearance other peaks at 588 m/z back to exit of the two chlorine atoms $[CuL_2]^+$ and 523 m/z due to L_2 $[C_{32}H_{24}N_6O_2]^+$.

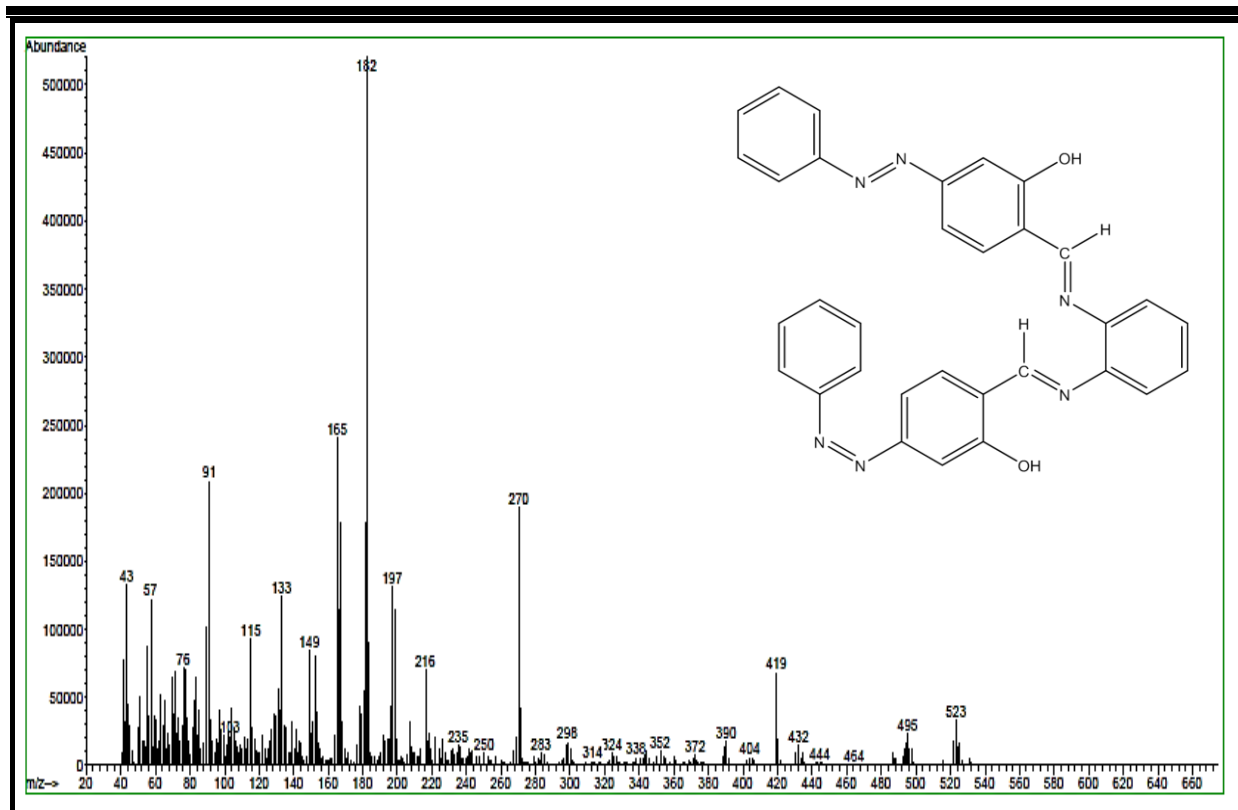
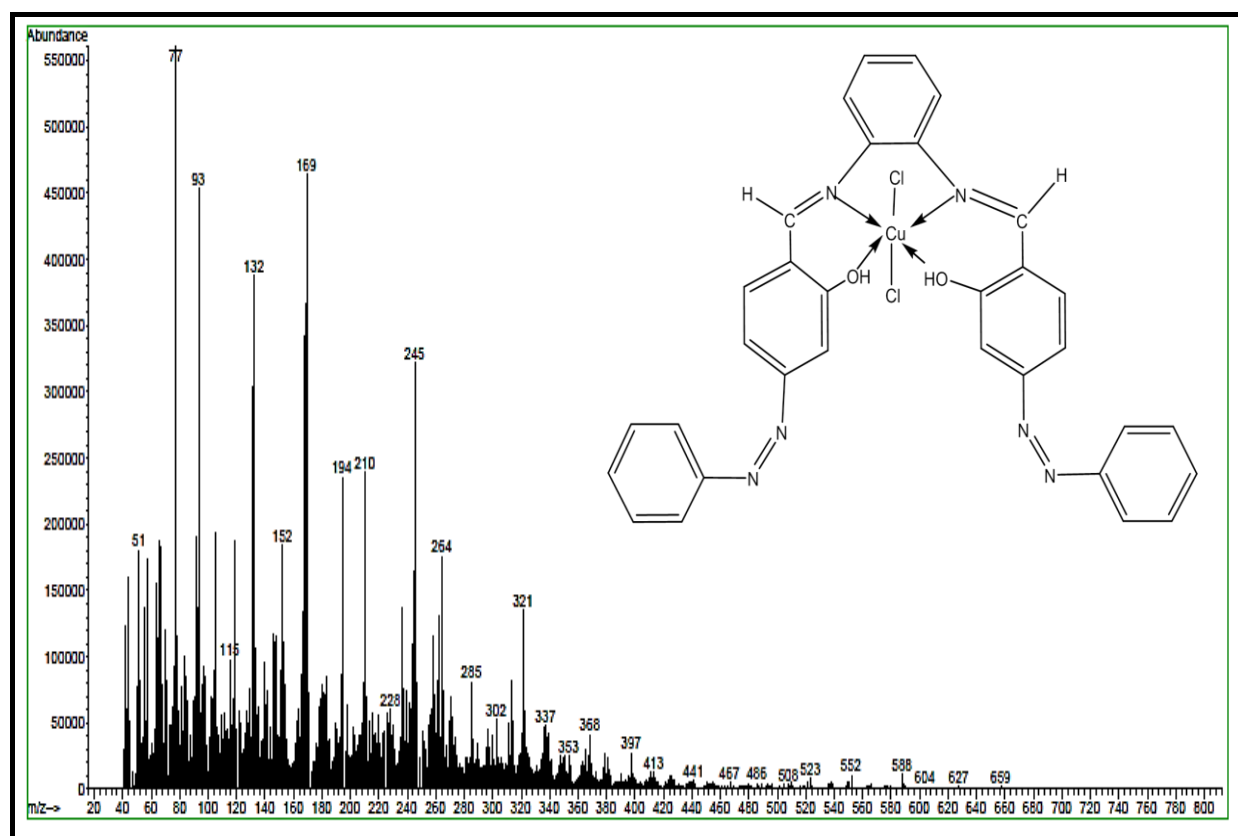
While the mass spectra of the complex $[CoL_2Cl_2]$ (figure 3.56) shows of molecular ion (M^+) at 656 m/z and appearance other peaks at 621m/z , 587 m/z

back to exit of the chlorine atoms $[\text{CoL}_2\text{Cl}]^+$, $[\text{CoL}_2]^+$ and 524 m/z due to L_2 $[\text{C}_{32}\text{H}_{24}\text{N}_6\text{O}_2]^+$.

Table (3.6): Includes the important peaks which attributed to the fragment of L_2

m/z	
523	$[\text{C}_{32}\text{H}_{24}\text{N}_6\text{O}_2]^+$
432	$[\text{C}_{26}\text{H}_{19}\text{N}_5\text{O}_2]^+$
419	$[\text{C}_{26}\text{H}_{19}\text{N}_4\text{O}_2]^+$
314	$[\text{C}_{20}\text{H}_{14}\text{N}_2\text{O}_2]^+$
298	$[\text{C}_{20}\text{H}_{13}\text{N}_2\text{O}]^+$
197	$[\text{C}_{12}\text{H}_9\text{N}_2\text{O}]^+$
182	$[\text{C}_{12}\text{H}_{10}\text{N}_2]^+$
133	$[\text{C}_7\text{H}_5\text{N}_2\text{O}]^+$
103	$[\text{C}_7\text{H}_5\text{N}]^+$
91	$[\text{C}_6\text{H}_5\text{N}]^+$
76	$[\text{C}_6\text{H}_4]^+$

Scheme (3.2): Mass spectra for the ligand L_2

Figure (3.54): Mass spectrum of the L_2 Figure (3.55): Mass spectrum of the complex $[CuL_2Cl_2]$

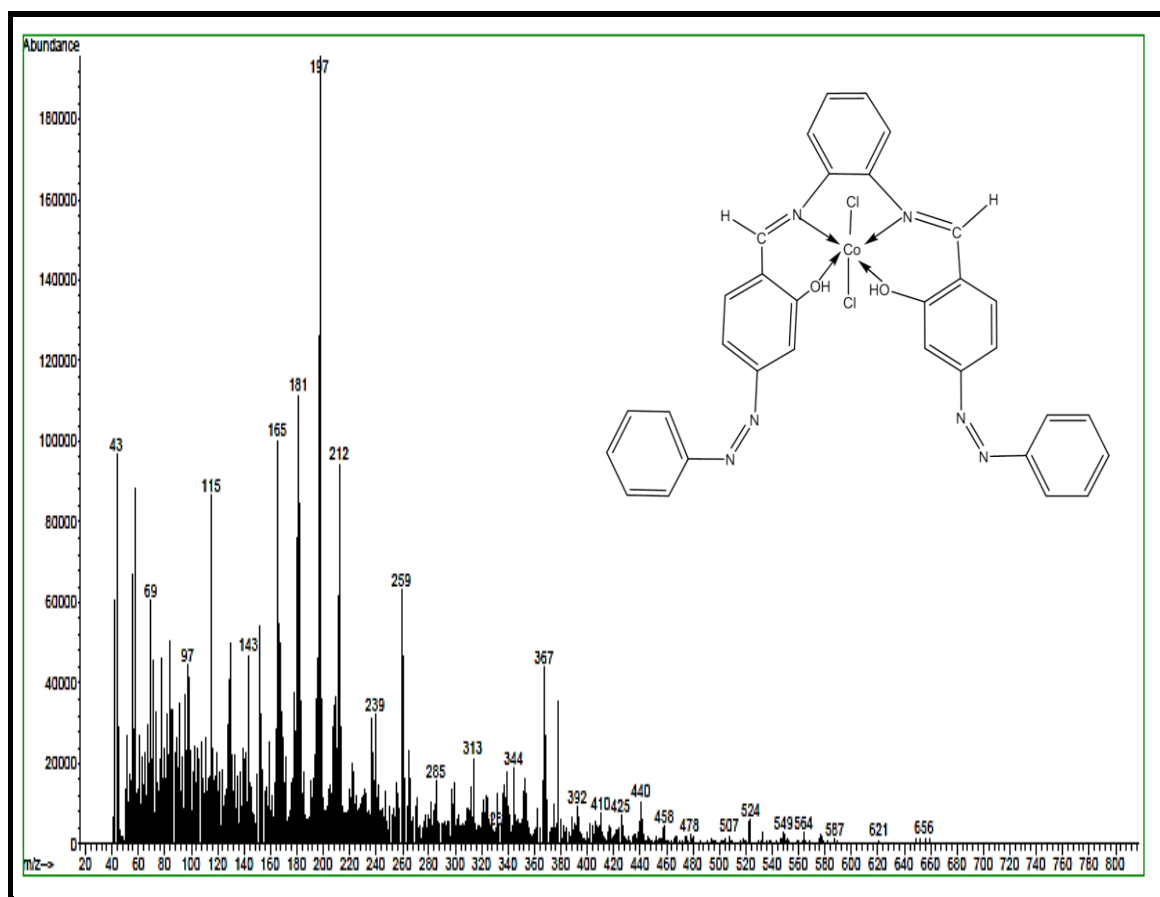
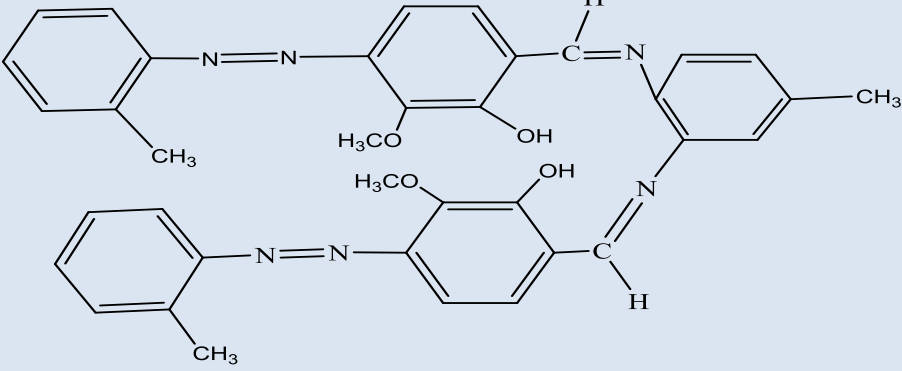


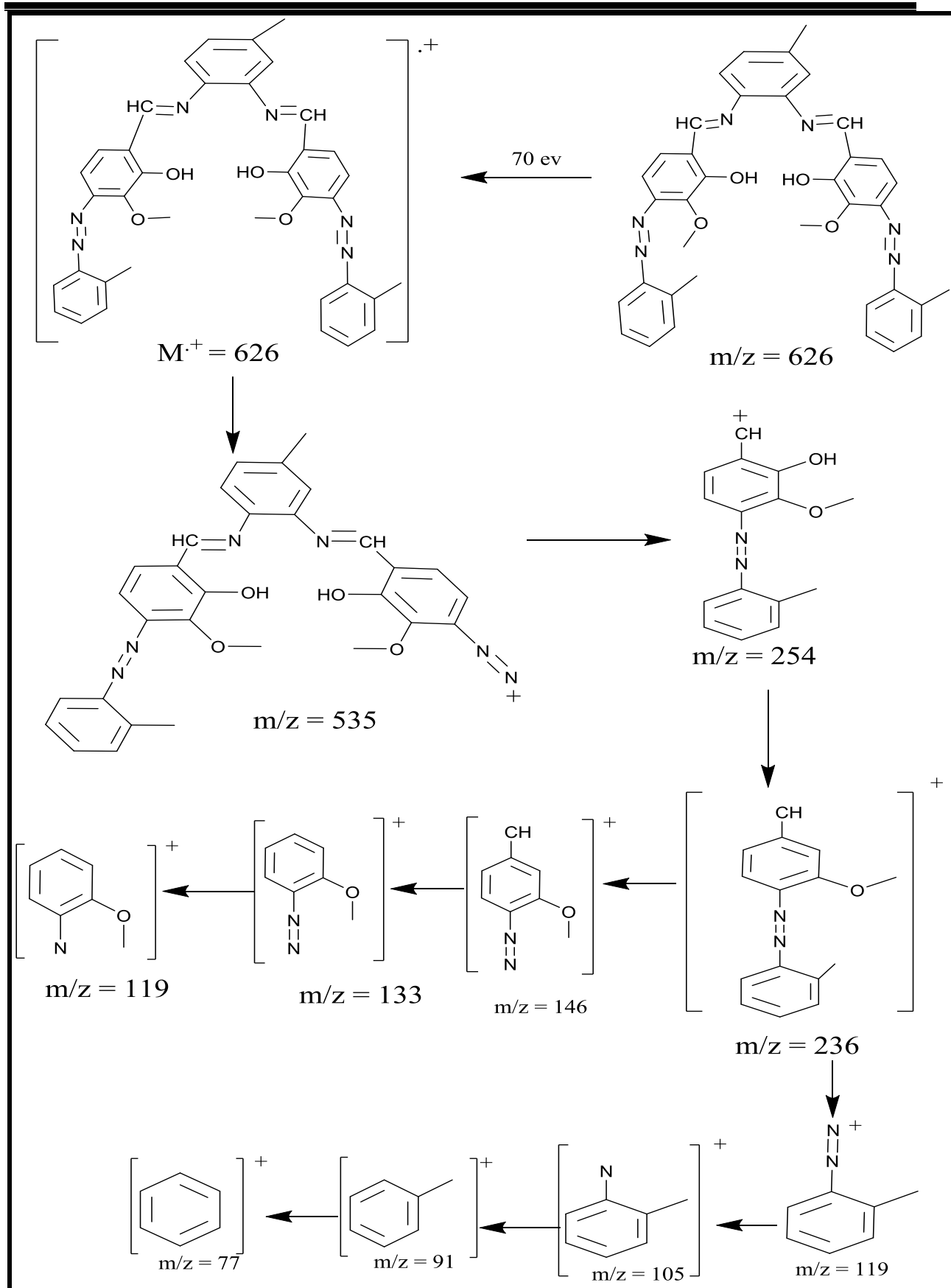
Figure (3.56): Mass spectrum of the complex $[\text{CoL}_2\text{Cl}_2]$

3.7.3: Mass spectra of L_3

Characterize mass spectra of the ligand appearance of molecular ion peak (M^+) at 626 m/z and the spectrum show a series of peaks (figure 3-57) at m/z = 535, 254, 236, 146, 133, 119, 105, 91 and 77 corresponding to $[\text{C}_{30}\text{H}_{27}\text{N}_6\text{O}_4]^+$, $[\text{C}_{15}\text{H}_{14}\text{N}_2\text{O}_2]^+$, $[\text{C}_{15}\text{H}_{13}\text{N}_2\text{O}]^+$, $[\text{C}_8\text{H}_6\text{N}_2\text{O}]^+$, $[\text{C}_7\text{H}_5\text{N}_2\text{O}]^+$, $[\text{C}_7\text{H}_7\text{N}_2]^+$, $[\text{C}_7\text{H}_7\text{N}]^+$, $[\text{C}_7\text{H}_7]^+$ and $[\text{C}_6\text{H}_5]^+$ respectively. They are listed in table (3-7). The mechanics of break- up are embedded in the scheme (3-3).

Table (3.7): Includes the important peaks which attributed to the fragment of L₃

m/z	
626	$[C_{37}H_{34}N_6O_4]^+$
535	$[C_{30}H_{27}N_6O_4]^+$
254	$[C_{15}H_{14}N_2O_2]^+$
236	$[C_{15}H_{13}N_2O]^+$
146	$[C_8H_6N_2O]^+$
133	$[C_7H_5N_2O]^+$
119	$[C_7H_7N_2]^+$
105	$[C_7H_7N]^+$
91	$[C_7H_7]^+$
77	$[C_6H_5]^+$

Scheme (3.3): Mass spectra for the ligand L_3

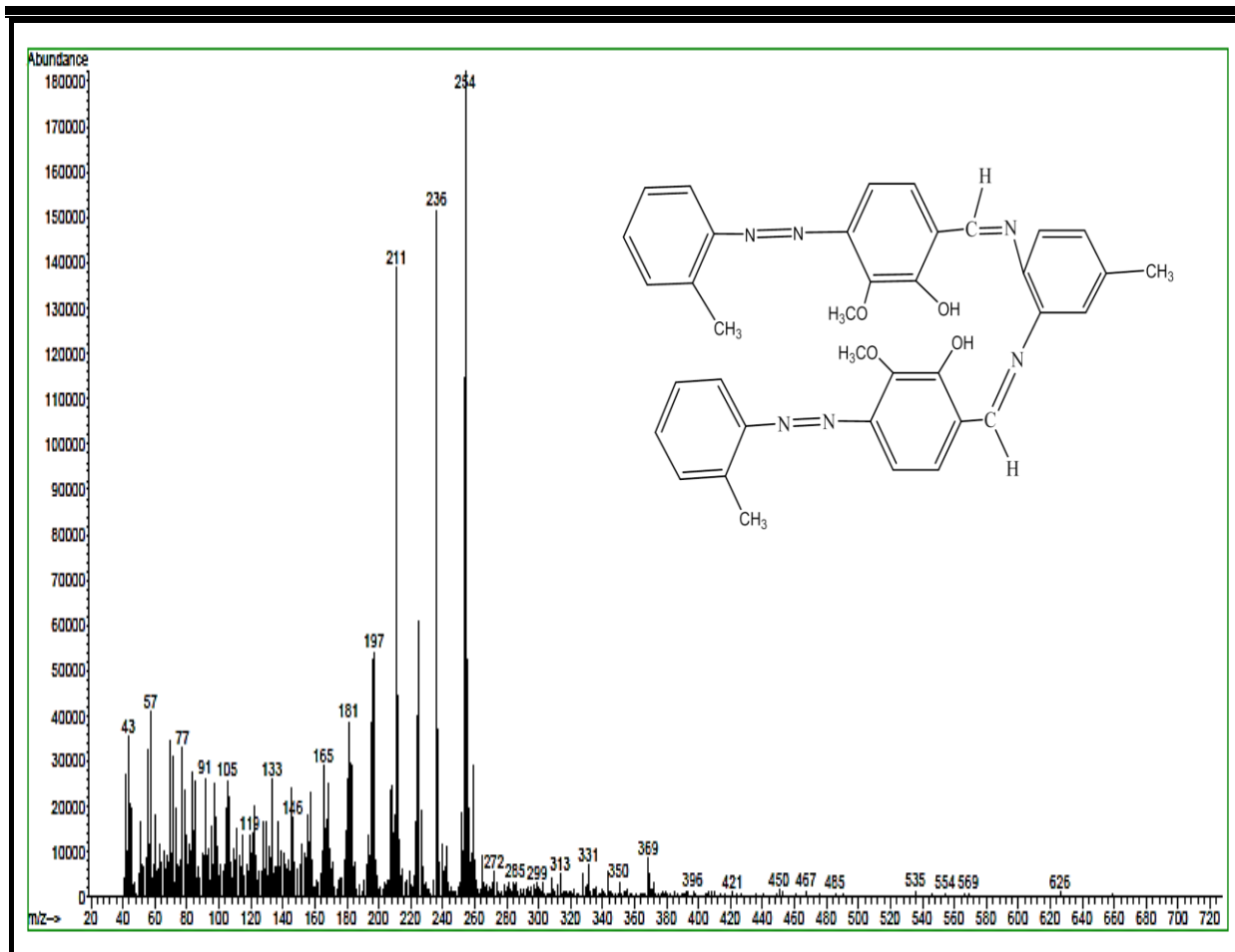


Figure (3.57): Mass spectrum of the L_3

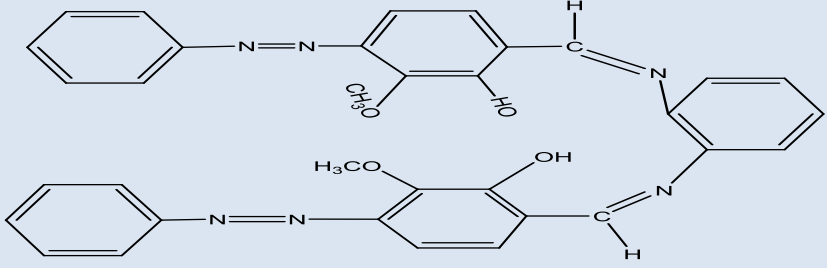
3.7.4 : Mass spectra of L_4 and their complexes

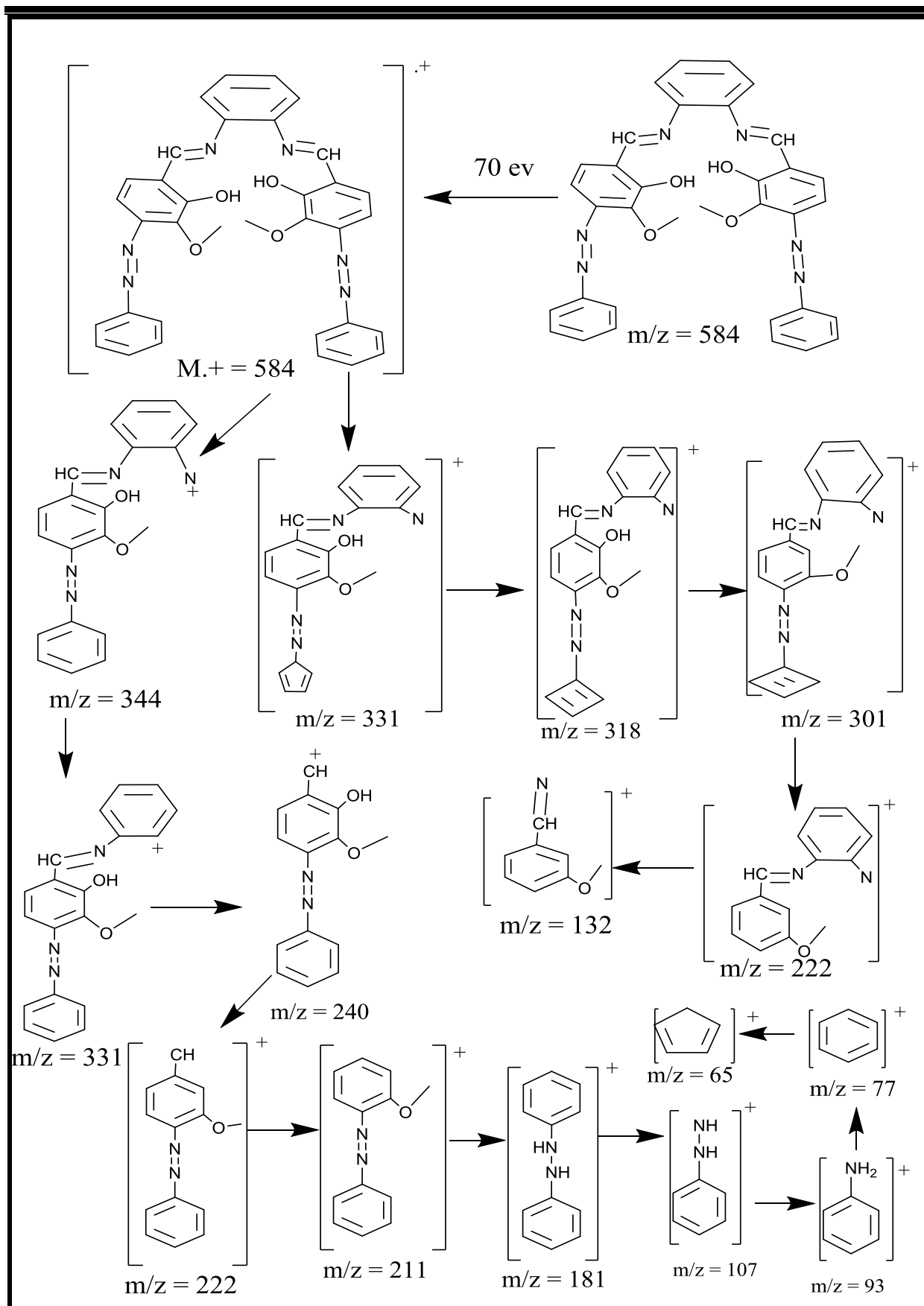
Characterize mass spectra of the ligand appearance of molecular ion peak (M^+) at 584 m/z and the spectrum show a series of peaks (figure 3.58) at m/z = 344, 331, 318, 301, 240, 222, 211, 181, 132, 107, 93, 77 and 65 corresponding to $[C_{20}H_{16}N_4O_2]^+$, $[C_{20}H_{16}N_3O_2]^+$, $[C_{18}H_{14}N_4O_2]^+$, $[C_{18}H_{13}N_4O]^+$, $[C_{14}H_{12}N_2O_2]^+$, $[C_{14}H_{11}N_2O]^+$, $[C_{13}H_{11}N_2O]^+$, $[C_{12}H_9N_2]^+$, $[C_8H_6NO]^+$, $[C_6H_7N_2]^+$, $[C_6H_7N]^+$, $[C_6H_5]^+$ and $[C_5H_5]^+$ respectively. They are listed in table (3.8). The mechanics of break- up are embedded in the scheme (3.4).

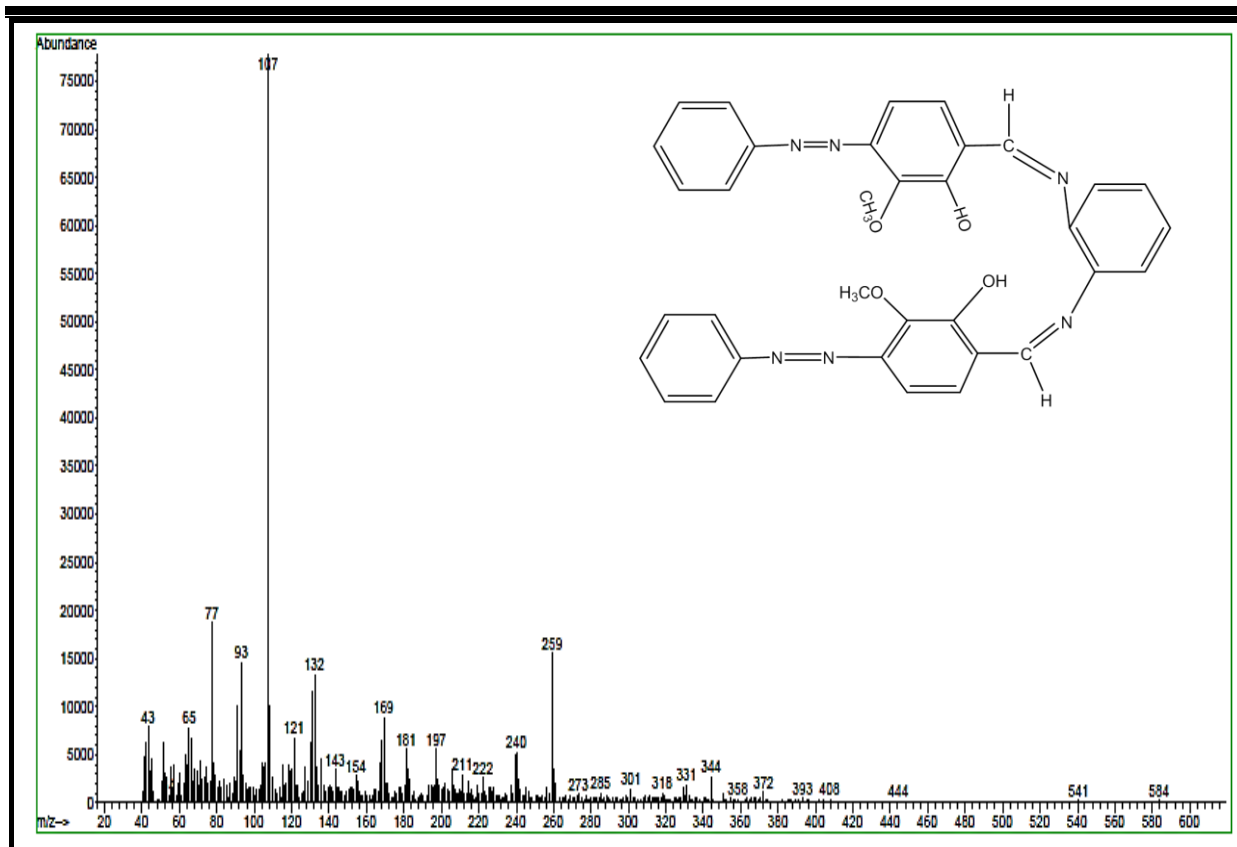
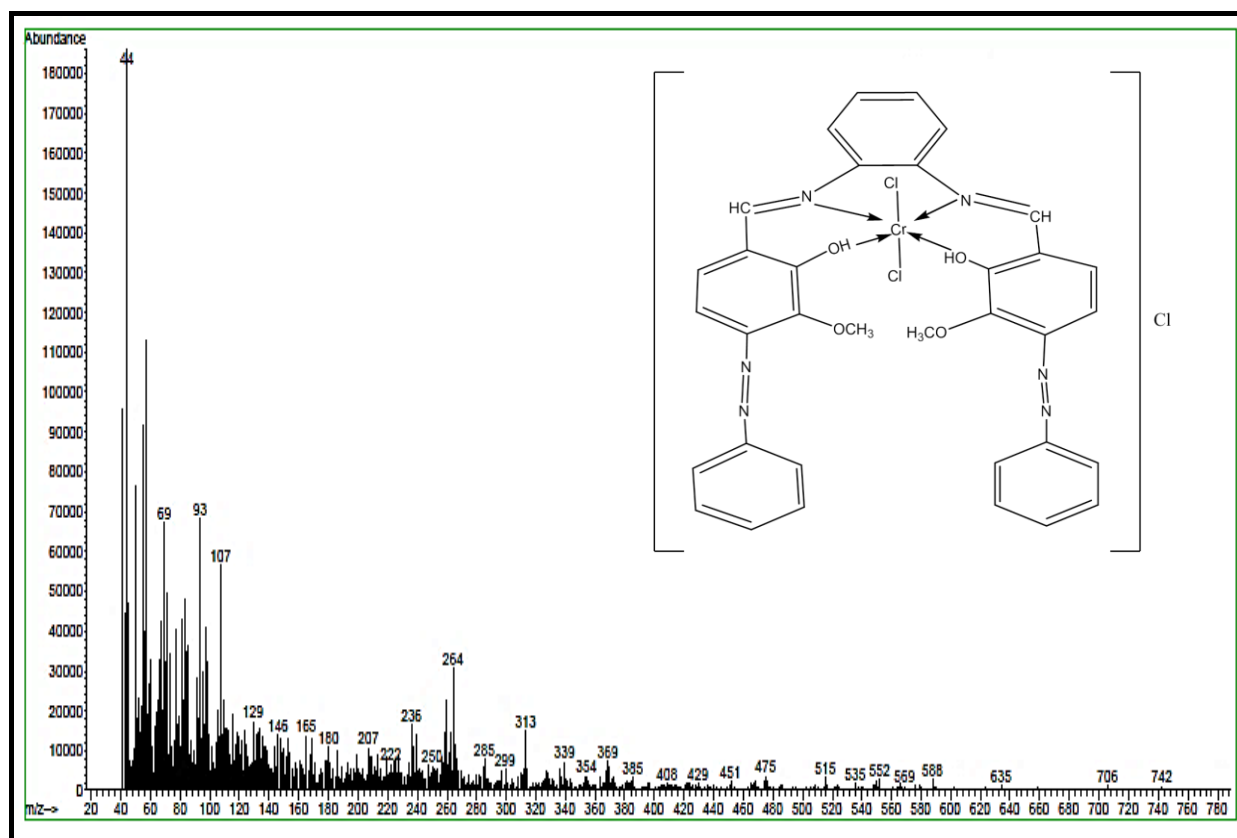
The mass spectra of the complex $[CrL_4Cl_2]Cl$ (figure 3.59) shows of molecular ion (M^+) at 742 m/z and appearance other peaks at 706 m/z , 635 m/z back to exit of the chlorine atoms $[CrL_4Cl_2]^+$, $[CrL_4]^+$.

While the mass spectra of the complex [CdL₄Cl₂] (figure 3.60) shows of molecular ion (M⁺) at 767 m/z and appearance other peaks at 732 m/z , 583 m/z back to exit of the chlorine atom [CdL₄Cl]⁺ and 583 m/z due to L₄ [C₃₄H₂₈N₆O₄]⁺.

Table (3.8): Includes the important peaks which attributed to the fragment of L₄

m/z	
584	[C ₃₄ H ₂₈ N ₆ O ₄] ⁺
344	[C ₂₀ H ₁₆ N ₄ O ₂] ⁺
331	[C ₂₀ H ₁₆ N ₃ O ₂] ⁺
318	[C ₁₈ H ₁₄ N ₄ O ₂] ⁺
301	[C ₁₈ H ₁₃ N ₄ O] ⁺
240	[C ₁₄ H ₁₂ N ₂ O ₂] ⁺
222	[C ₁₄ H ₁₁ N ₂ O] ⁺
211	[C ₁₃ H ₁₁ N ₂ O] ⁺
181	[C ₁₂ H ₉ N ₂] ⁺
132	[C ₈ H ₆ NO] ⁺
107	[C ₆ H ₇ N ₂] ⁺
93	[C ₆ H ₇ N ₁] ⁺
77	[C ₆ H ₅] ⁺
65	[C ₅ H ₅] ⁺

Scheme (3.4): Mass spectra for the ligand L_4

Figure (3.58): Mass spectrum of the L_4 Figure (3.59): Mass spectrum of the complex $[CrL_4Cl_2]Cl$

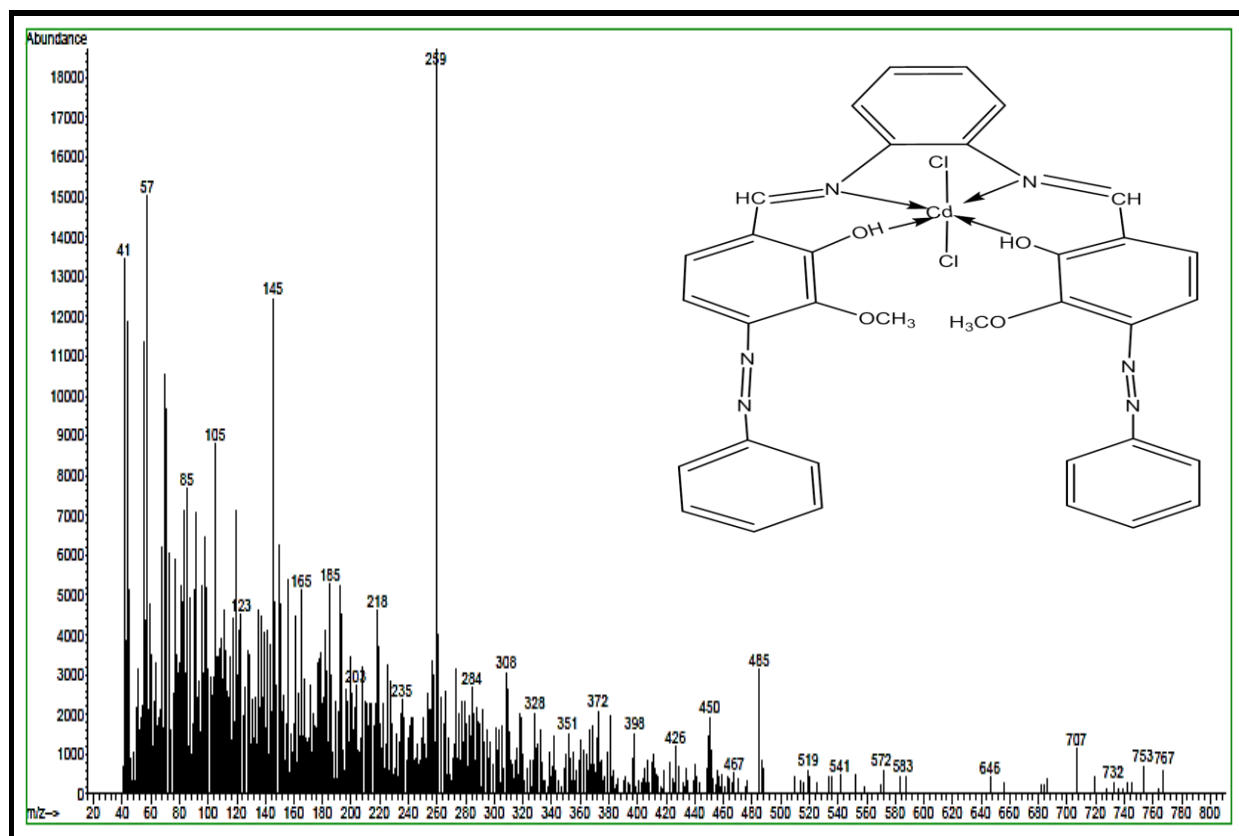


Figure (3.60): Mass spectrum of the complex $[CdL_4Cl_2]$

3-8: Molar Electrical Conductivity

The electrical conductivity is regarded as one of the important and simple means for knowing the ionic formula of the compounds which are complex in their solutions and the degree of electrical conductivity is direct proportional with the number of ions which are released by the complex in the solution. It takes low values, when the complex does not have any ionic feature⁽²¹²⁾. The water is not favored to be used as a solvent in the process of measuring the molar electrical conductivity for coordinative complexes because of some complexes sometimes hydrolysis or difficult dissolved in water, But the organic solvents are often used, such as methyl cyanide, nitro methane, dimethyl formamide, dimethyl sulfoxide and other, where the solvent inactive for the complexes have a dielectric constant and low viscosity⁽²¹³⁾.

Definition molar conductivity 1 cm^3 of solution that contains 1 mole of the solute and the molar conductivity can be counted by the following relations.

L complex = A solution – A solvent

K = L complex × K cell

$\Lambda_m = 1000\text{ K/C}$

K cell = cell constant (cm^{-1})

L complex = complex conductivity (S)

A solution = solution conductivity (S)

A solvent = solvent conductivity (S)

K = specific conductance (S.cm^{-1})

C = molar concentration (mole. cm^{-3})

Λ_m = molar conductivity ($\text{S.cm}^2.\text{mole}^{-1}$)

Table (3.9): The values of molar conductivity Λ_m complexes ligand L_2 in solvent DMSO concentration of 10^{-3} M at a temperature of 298 K

No.	Complexes	$\Lambda_m (\text{s.cm}^2.\text{mole}^{-1})$ in DMSO	Electrolyte Type
1	$[\text{CuL}_2\text{Cl}_2]$	17	Non Electrolyte
2	$[\text{CoL}_2\text{Cl}_2]$	16	Non Electrolyte

Table (3.10): The values of molar conductivity Λ_m complexes ligand L_4 in solvent DMSO concentration of 10^{-3} M at a temperature of 298 K

No.	Complexes	$\Lambda_m (\text{s.cm}^2.\text{mole}^{-1})$ in DMSO	Electrolyte Type
1	$[\text{CrL}_4\text{Cl}_2]\text{Cl}$	34	1:1
2	$[\text{CdL}_4\text{Cl}_2]$	15	Non Electrolyte

It was measured molar conductivity for solid complexes solutions of ions Cu(II) and Co(II) ions with L_2 and Cr(III) and Cd(II) ions with L_4 concentration of $10^{-3}M$ dissolved in dimethylsulfoxide (DMSO) each separately at a laboratory temperature, has been used distilled water as a reference. The results are included in table (3.9) and table (3.10) were found from the conductivity values of the Cu(II), Co(II) and Cd(II) complexes behave neutral compounds (non electrolytic) the lack of any adjective ionic but Cr(III) complex with L_4 behaves the behavior of ionic compound and its conductivity is 1:1 depending on the chloride ions outside. To make sure there is no chloride outside the coordination sphere by adding aqueous solution of silver nitrate ($AgNO_3$) to complex solution (complex dissolved in DMSO), where noted not appearance precipitate or turbidity the solution, this refers to the absence of chloride ion outside the coordination sphere while if the chloride out of the coordination sphere white precipitate it will form, and turbidity increased by increasing the number chloride atoms outside of the coordination sphere as counter ions. The obtained results were appropriate with the molecular formula and stereochemistry proposed of the prepared complexes.

Cloud Point Extraction

In this study, unprecedented trends for the analytical separation and preconcentration by using the cloud-point extraction (CPE) methodology, three chelating agents were used for formation of ion association complexes with Cd(II), Cu(II) and Co(II) ions. L₁ with Cd(II), L₂ with Cu(II) and L₃ with Co(II). These reagents were synthesized in our laboratory.

3.9: Cloud point extraction of cadmium(II)

3.9.1: Calibration curve of determination cadmium ion

A calibration curve was previously constructed for each experiments in this study in order to determine the quantity of Cd(II) ion. The absorbance was recorded in the concentration range (1.6) $\mu\text{g}.\text{ml}^{-1}$ of Cd(II) at λ_{max} of 228.80 nm by flame atomic absorption spectrophotometer. The calibration curve was constructed by plotting values of absorbance versus concentration as shown in figure (3.61).

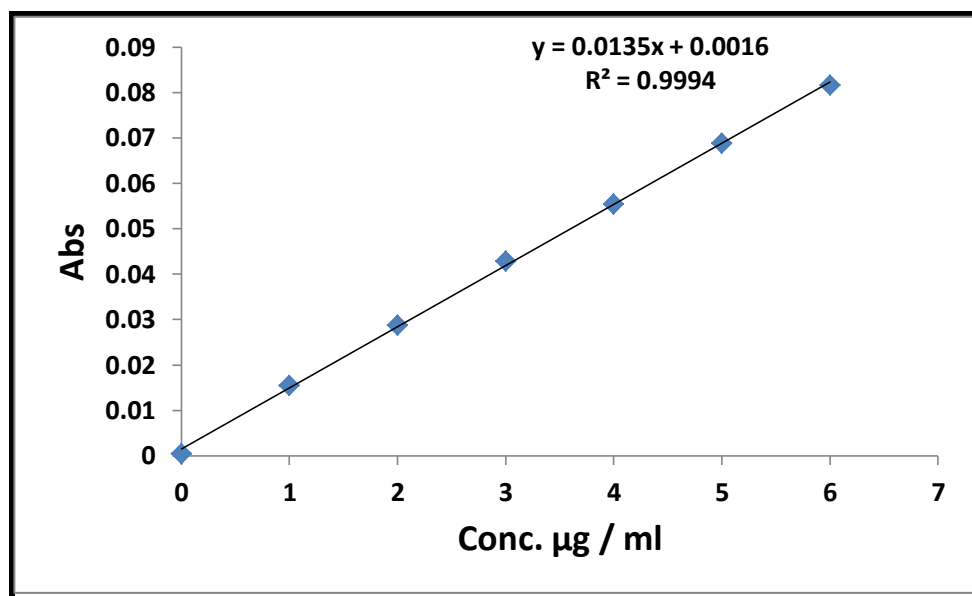


Figure (3.61): Calibration curve of cadmium (II) ion

3.9.2: Optimization of CPE Procedure

The effects of several experimental parameters which impact the CPE efficiency were carried out by classical optimization (one-variable-at-a-time). In this approach, we observe the effect of one factor at a time (OVAT) on an experimental response. While only one factor is changed, others are kept at a constant level. Although, the “optimization” performed by OVAT⁽²¹⁴⁾ does not ensure at all that the real optimum will be conformed, but it would be valid only if the variables to be optimized would be totally independent from each other (i.e. no interactive effects among the variables). Nevertheless, the classical optimization certainly leads at least to an improvement of the analytical method. Inasmuch as the extraction efficiency of the CPE depends on dual factors, some of regarding the prior formation of a complex with sufficient hydrophobicity and the other for the formation of micelles to obtain the desired separation and preconcentration. Consequently, the effects of reagent concentration, HCl concentration, incubation time, equilibration temperature, Triton X-100 volume were selected in this study.

3.9.2.1: Effect of reagent (L_1) Concentration

The effect of the concentration of reagent (L_1) was investigated by taking 10 ml solution containing $3\mu\text{gml}^{-1}$ of Cd(II), 0.5 ml of 1% Triton X-100, 1ml of 0.1M HCl and varying concentration of (1×10^{-4} , 5×10^{-4} , 1×10^{-3} , 5×10^{-3} , 1×10^{-2} , 5×10^{-2} M) of reagent (L_1), followed CPE procedure (2.13). The results are shown in figure (3.62) .

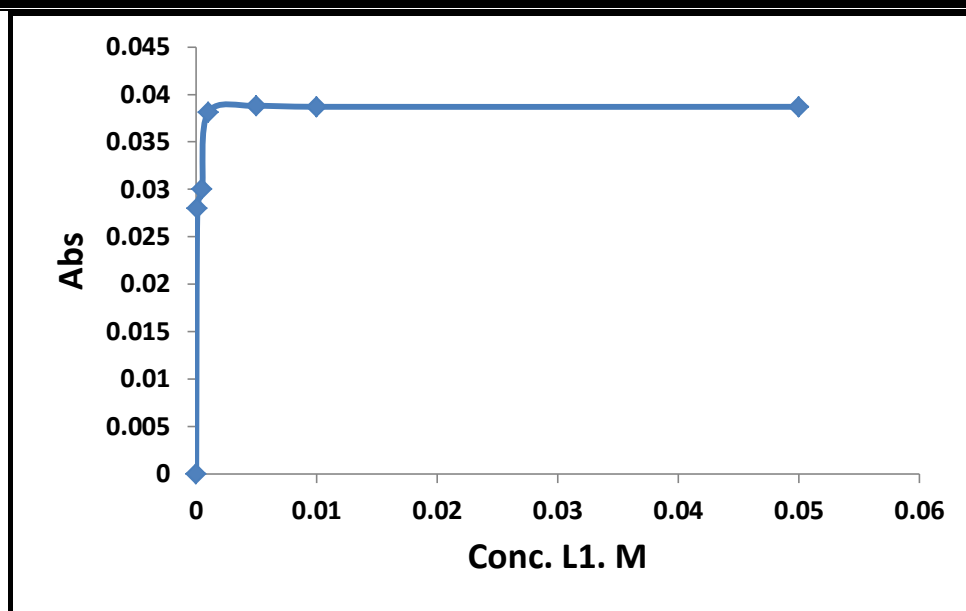
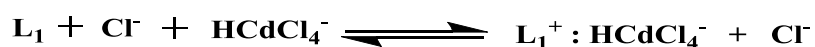


Figure (3.62): Effect L_1 concentration on the absorbance values

The absorbance increases linearly with an increase in L_1^+ concentration up to 5×10^{-3} M which was certainly sufficient for ion-association complex formation in the extracted medium. After this concentration, the absorbance remained constant indicating that the extraction of the complex in micelles medium was independent of further increase in the concentration of L_1^+ . Therefore, a concentration of 5×10^{-3} M of L_1^+ was decided to be used as optimum the provides a high distribution ratio (D) with highly stable ion pair complex formation extractable into surfactant. The distribution ratio (D) to obtain from the reminder quantity of Cd(II) as $CdCl_4^{2-}$ and amount of transferred Cd(II) as $CdCl_4^{2-}$ to form ion-pair complex into surfactant .

At lower than 5×10^{-3} M, unstable ion-pair complex was formed which leads to depress the extraction into surfactant because the complexation reaction is not sufficient to reach an ideal thermodynamic equilibrium for complex formation because the rate of dissociation becomes more than the rate of association⁽¹⁸⁶⁾. Therefore, we chose a concentration of 5×10^{-3} M of L_1 as optimal in this work.



3.9.2.2: Effect of HCl Concentration

Because the HCl concentration plays an important role in metal anion CdCl_4^{2-} complex formation with reagent and subsequent extraction, the effect of HCl concentration was studied by measuring the absorbance of solutions containing $3\mu\text{g.mL}^{-1}$ cadmium as CdCl_4^{2-} , $5 \times 10^{-3}\text{M}$ reagent(L_1) and 0.5 ml of 1% (w/v) Triton X-100 with various concentrations of hydrochloric acid (0.05-2M), according to general CPE procedure (2.13) as shown in figure (3.63).

The results reveal that the absorbance is almost constant in the HCl concentration range of (0.5-1)M. At lower than 0.5M HCl, the absorbance depression occurs due to incomplete ion pair complex formation between CdCl_4^{2-} and L_1 , while the above 0.5M HCl the absorbance slightly decreases because of the instability of ion pair complex and less micelles formation with surfactant Triton X-100. Thus 0.5M of HCl was selected as optimum value throughout the experiment⁽¹⁸⁶⁾.

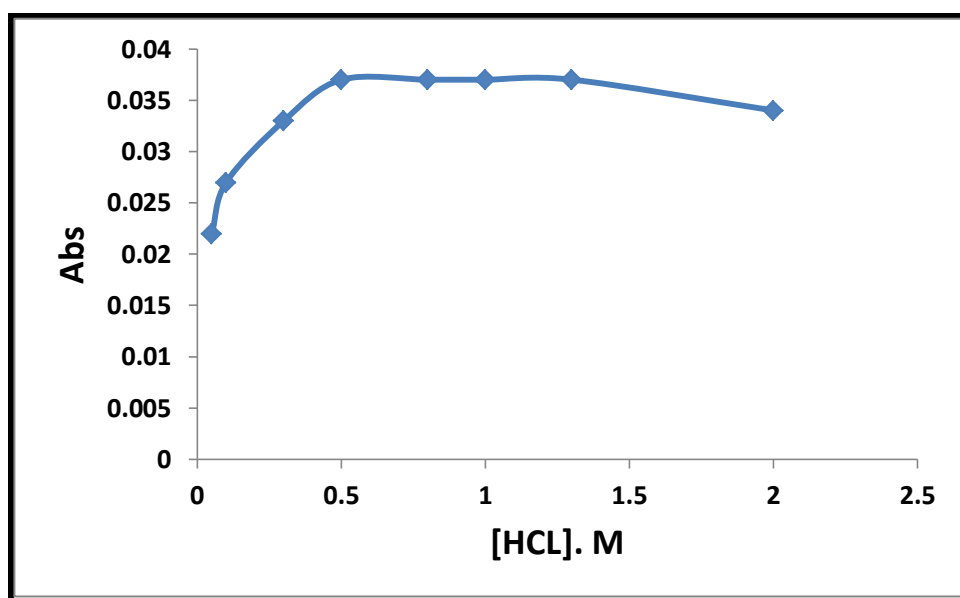
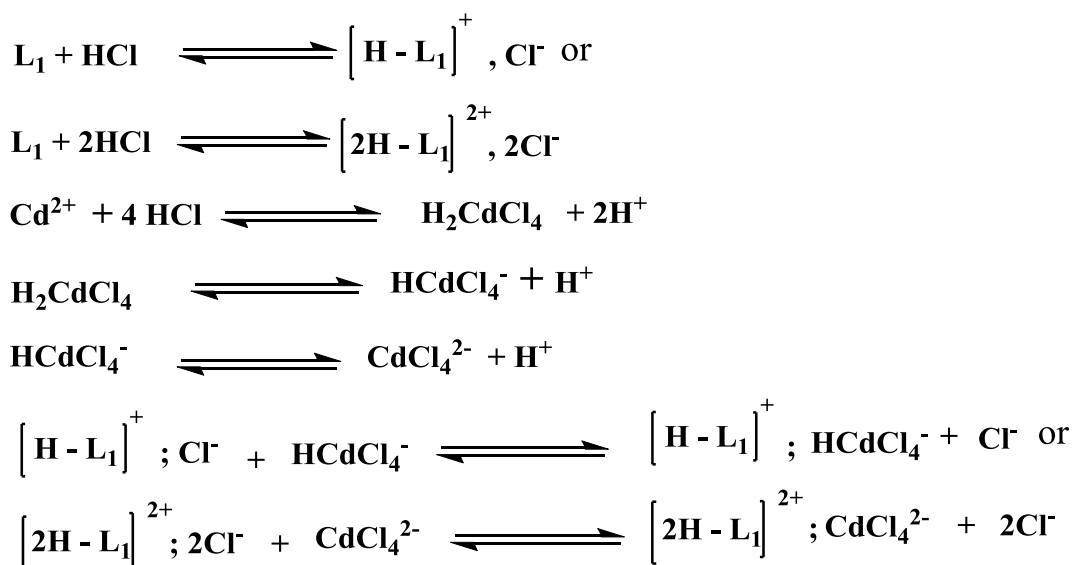


Figure (3.63): Effect of HCl concentration on CdCl_4^{2-} complex formation with L_1 by CPE

The results show that 0.5M HCl more favourable for the extraction of cadmium as anion complex CdCl_4^{2-} with reagent (L_1) in the form of ion association complex, giving higher absorbance for complex extracted to micelles layer. It is worth mentioning the transformation of cadmium cation to anion complex CdCl_4^{2-} to submit to thermodynamic equilibria to the formation of high concentration and stable anion complex according to equilibriums below:-



This thermodynamic equilibrium depends on chloride ion concentration in aqueous solution and concentration of anion complex (CdCl_4^{2-}) as well the high stability effect on the extraction efficiency by cloud point extraction method. At concentration of HCl less than 0.5M, it cannot be allowed to reach the thermodynamic equilibrium, which is not giving stable and suitable concentration of CdCl_4^{2-} for efficiency extraction, but more than 0.5M of HCl the effect leads to depress anion complex formation and stability as well as decrease absorbance of ion pair complex extracted to cloud point and distribution ratio (D) according to mass action law and Le Chatlier principle⁽²¹⁵⁾.

3.9.2.3: Effect of Incubation Time

The effect incubation time on extraction is an important parameter in CPE process for complete reaction to achieve easy phase separation and the of Cd(II) preconcentration to be as efficient as possible. The effect of time on the extraction as CdCl_4^{2-} in the range of (10 - 45) min was investigated by taking 10ml aqueous solution containing $3\mu\text{gml}^{-1}$ Cd^{2+} , 0.5 M HCl, 5×10^{-3} M (L_1) ,0.5ml of 1% (w/v) Triton X-100, followed CPE procedure (2.13). The results are shown in figure (3.64).

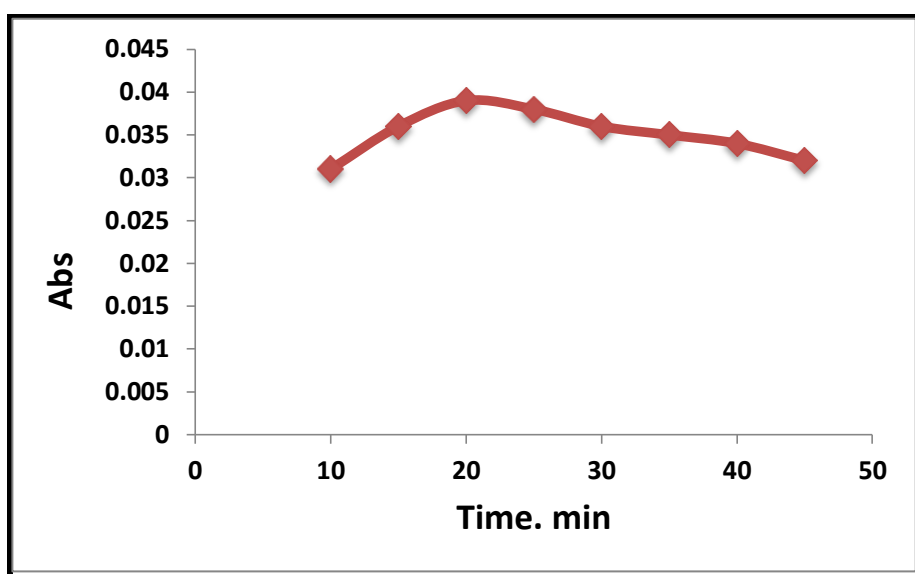


Figure (3.64): Effect of time on absorbance

It showed that the maximum time of incubation was observed at 20 min and after this value resulted in poor quantitative extraction occurs, probably due to thermal instability of the complex. Therefore, an incubation time at 20 min was selected as optimal with a highly stable ion pair complex formation extractable into surfactant⁽¹⁸⁶⁾. Any time less than optimum not allow reaching kinetic equilibrium for aggregation micelles, but the time more than optimum value giving increasing in kinetic energy which is effect to increase diffusion of micelles⁽²¹⁶⁾.

3.9.2.4: Effect of equilibrium temperature

The effect of the equilibrium temperature was examined by taking 10 ml aqueous phase containing of $3\mu\text{gml}^{-1}\text{Cd (II)}$ as CdCl_4^{2-} and keeping other factors at optimum conditions. The temperature was varied from 30 °C to 90 °C for 20 min followed the general CPE procedure(2-13). Preliminary experiments indicated that at temperature below 70 °C, no concrete phase separation (i.e. two phases weren't formed) was observed due to very low number of micelles formed which cannot be able to encompass the ion-pair association complex at this temperature. But, at 70 °C, an excellent phase separation has taken place because of high number of micelles formed at CPT leading the entire transference of the hydrophobic complex into surfactant-rich phase that maximizes the sensitivity⁽¹⁸⁶⁾ as shown in figure (3.65).

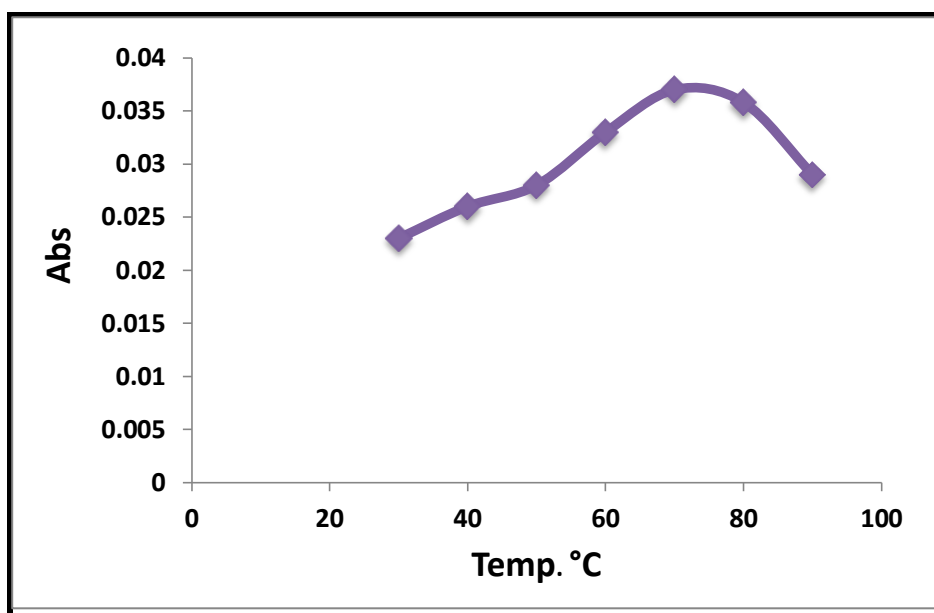


Figure (3.65): Effect of temperature on complex formation by CPE

3.9.3: Thermodynamic Study

The equilibrium extraction constants (K_{ex}) and thermodynamic parameters for the extraction of $[L_1^+;HCdCl_4^-]$ complex during the cloud point using Triton X-100 as a mediated extracting agent at various temperatures were determined. Depending on different operation temperatures and the distribution of target species between two phases via calculating the distribution ratios (D), the equilibrium extraction constants (K_{ex}) can be calculated and subsequently thermodynamic parameters for the extraction of the complex system at various temperatures were also determined. The equilibrium constants (K_{ex}) at the selected temperature were calculated from equation mentioned in (2.14.4) and the results are presented in table (3.12) and shown in figure (3.66).

Table (3.11): Variation of temperature for the extraction cadmium by L_1 on distribution ratio D.

T / °C	T / °K	Absorbance	D
30	303	0.023	1.089
40	313	0.026	1.477
50	323	0.028	1.929
60	333	0.033	3.386
70	343	0.037	7.475
80	353	0.036	5.466
90	363	0.029	2.112

Table (3.12): Variation of equilibrium constant with temperature during CPE for extraction Cd(II) with (L_1)

T°C	30	40	50	60	70
T°K	303	313	323	333	343
1/ T *10 ⁻³	3.300	3.195	3.096	3.003	2.915
D	1.089	1.477	1.929	3.386	7.475
K _{ex} * 10 ⁸	0.171	0.274	0.424	1.113	4.746
Log K _{ex} *10 ⁸	7.233	7.438	7.627	8.046	8.676

Thermodynamic parameters were also calculated from the relationships mentioned in (2.14.4). The results are summarized in table (3.13).

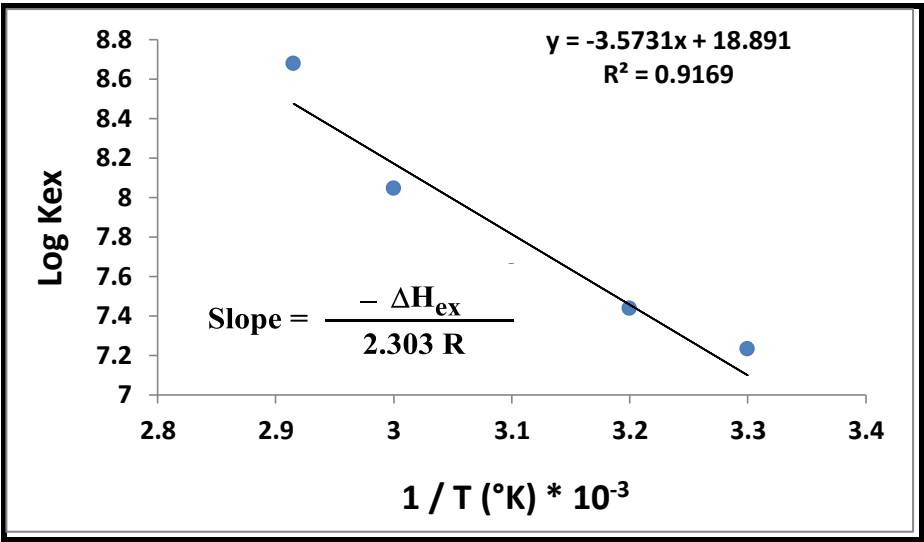


Figure (3.66): Effect of the equilibration extraction temperature on ion pair complex formation

Table (3.13): Thermodynamic parameters for the extraction of Cd ion-pair complex by CPE

T°K	ΔH_{ex} (KJ mol ⁻¹)	ΔG_{ex} (KJ mol ⁻¹)	ΔS_{ex} (Jmol ⁻¹)
303	0.0684	-41.958	138.701
313		-44.569	142.612
323		-47.166	146.237
333		-51.298	154.253
343		-56.974	166.304

The value of enthalpy change (ΔH_{ex}) was obtained from the plot of $\log K_{\text{ex}}$ versus $1/T$ (figure 3.66) and found to be $0.0684 \text{ kJ mol}^{-1}$ while the values of Gibbs free energy (ΔG_{ex}) were obtained at different temperatures. Once these two parameters are obtained, the values of entropy change (ΔS_{ex}) were also calculated and all the data are shown in (table 3.13). The results revealed that the extraction

of ion-pair complex is easy and thermodynamically favourable and resulting in increasing the phase-volume ratio thus extraction efficiency .

The extraction process is endothermic in nature, which is also verified from the positive value of ΔH_{ext} in (table 3.13). It was also noted from (table 3.13) that the values of ΔG_{ex} approximately increased with temperature. The negative values of ΔG_{ex} indicate that the ion-pair complex solubilisation process is also spontaneous and thermodynamically favourable^(216,217). The reaction of extraction by CPE is endothermic reaction and positive value of ΔS_{ex} means the reaction is entropic in region .

3.9.2.5: Effect of Triton X-100 Volume

A successful cloud point extraction procedure should maximize the extraction efficiency by minimizing the phase volume ratio ,which improve its concentrating capability. The effect of Triton X-100 concentration was investigated between (0.1-1.5)ml of triton X-100 (1% w/v) in 10ml aqueous solution containing $3\mu\text{gml}^{-1}$ Cd^{2+} , 0.5 M HCl and 5×10^{-3} M reagent and subjected to general CPE procedure (2.13). The results are shown in figure (3.67). Triton X-100 was chosen for the surfactant-rich phase due to its high extraction efficiency, adequate cloud-point temperature, and high viscosity of the surfactant-rich phase, which makes the phases separation easy without the need of centrifugation and cooling steps. The results have shown that the maximal absorbance signal were achieved when the volume of 1% (w/v) Triton X-100 at 0.9 ml (figure 3.67) and the absorbance subsequently decreased sharply because of the increment in the overall analyte volumes and viscosity of surfactant phase leading to poor sensitivity.

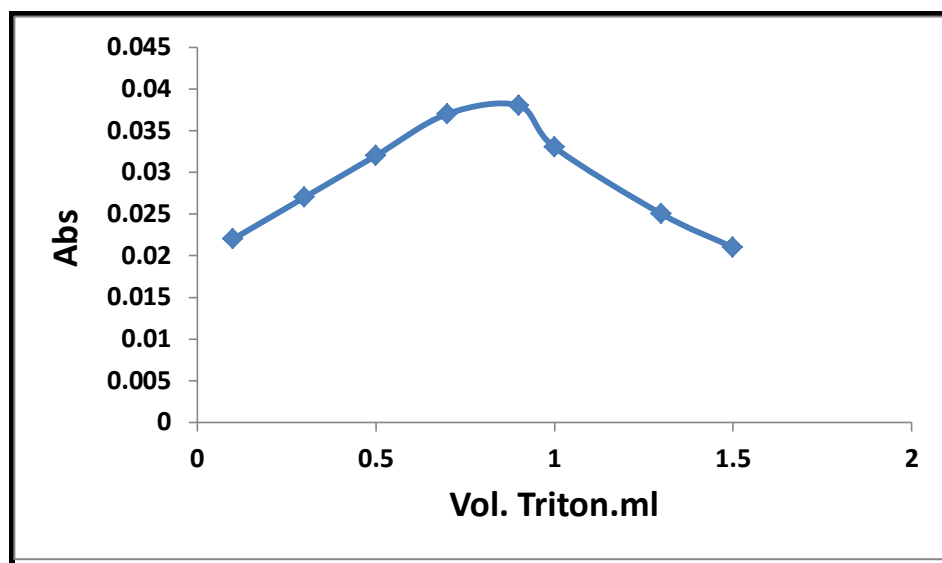


Figure (3.67): Effect of Triton (X-100) amount on absorbance

It can be concluded that at optimum amount, it means that thermodynamically plays a major role on extraction efficiency and this helps to reach favourable thermodynamic equilibrium for the extraction by giving a small viscous surfactant layer which can be extracted a high concentration of complex with high stability. At less than optimum of TX-100, it cannot be allowed to reach high stable thermodynamic equilibrium, while at concentration more than optimal, the effect leads to the deviation of the equilibrium due to the diffusion of micelles. Consequently, the optimum volume of 1% (w/v) Triton X-100 at 0.9ml was selected for optimal absorbance to achieve the highest possible extraction efficiency and pre-concentration factor^(186,218).

3.9.4: Stoichiometric Determination of Complex (Slope Analysis Method)

The stoichiometric determination for the reaction of CdCl_4^{2-} anion with L_1^+ organic reagent to form an ion-association complex was obtained according to the procedure mentioned in (2.16) by the dependence of $\log D = f(\log C_{\text{L}_1})$. The results are previously presented in shown in table (3.14) and figure (3.68).

Table(3.14): Effect of L_1 concentration on extractability of Cd(II)

$[L_1]$	Absorbance	D	Log D
1×10^{-4}	0.028	1.780	0.250
5×10^{-4}	0.030	2.448	0.389
1×10^{-3}	0.038	9.101	0.959
5×10^{-3}	0.039	10.364	1.016

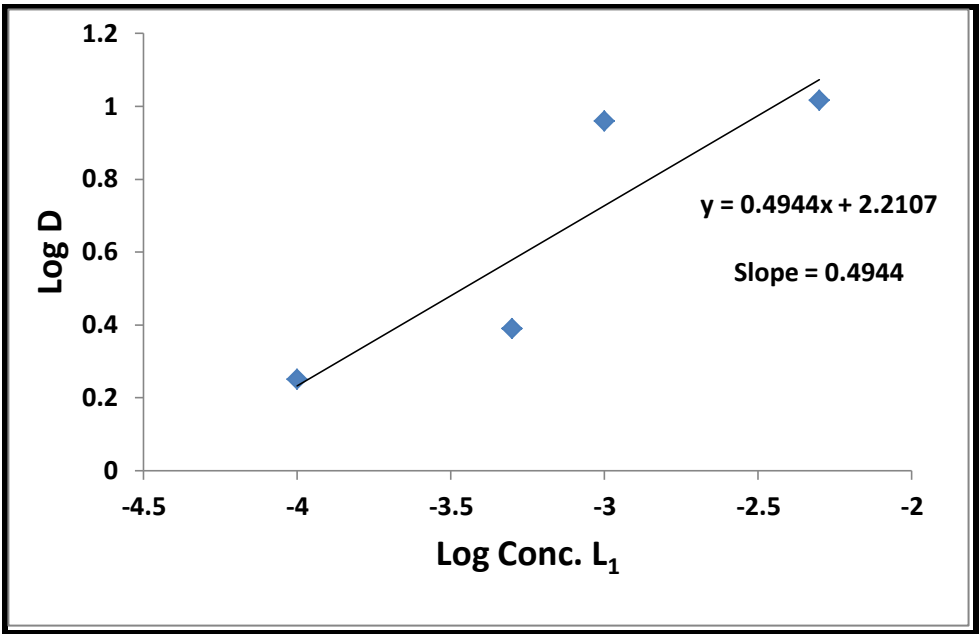


Figure (3.68): Slope analysis method: $\log D = f(\log C_{L1})$

The results revealed that the slope of the figure (3-68) in $\log D = f(\log C_{L1})$ coordinates is nearly equal to less than one (0.4944) , indicating that the ion pair complex with $[L_1^+; HCdCl_4^-]$ was most probably as1:1 ratio extracted into the surfactant-rich phase as shown in the following complexation reaction⁽²¹⁹⁾ .



3.9.5: Calibration Curve of Cd(II) using CPE

Calibration curve was obtained by plotting the absorbance versus the concentration of Cd^{+2} via the processing 15ml standard solutions containing $(0.01-1.3) \mu\text{g}.\text{ml}^{-1}$ of Cd^{2+} as CdCl_4^{2-} under the established optimized conditions from concentration of the reagent, concentration of HCl, Triton X-100, time and temperature. The absorbance of final solution was measured by FAAS. The results were shown in figure (3.69). Very good linearity was obtained in the concentration range of $(0.01-1.3) \mu\text{gml}^{-1}$ for Cd with coefficient of determination (R^2) of 0.9994.

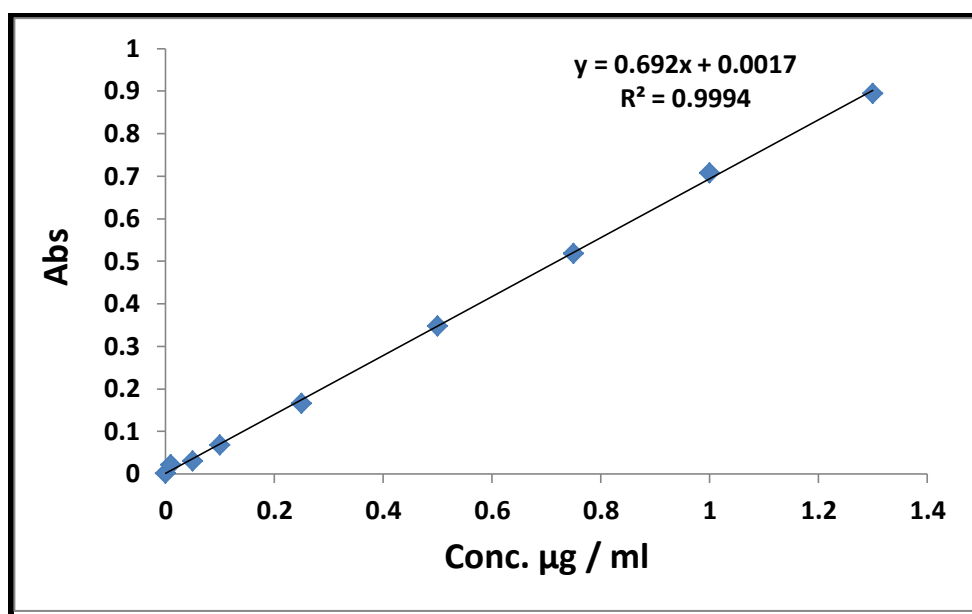


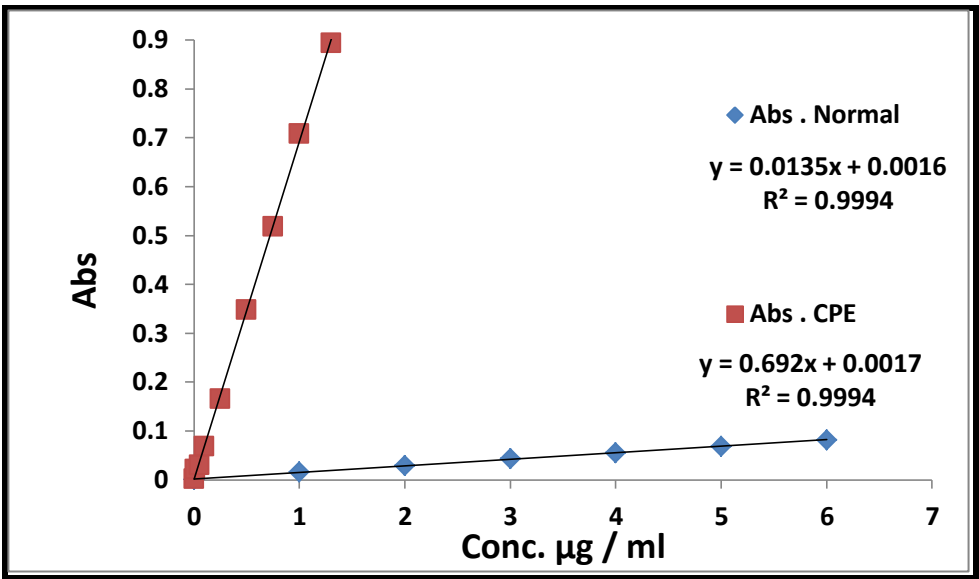
Figure (3.69): Calibration Curve for determination of cadmium ion using CPE-FAAS

3.9.6: Validation of Method and Calibration Curve of Cadmium Ion

Under the optimum conditions, performance characteristics were obtained by processing standard solutions of Cd^{2+} and the data is illustrated in table (3.15). Calibration function were linear at least up to $(0.01\text{-}1.3\mu\text{g.ml}^{-1})$. The limit of detection (LOD) of a method is the lowest analyte concentration that produces a response detectable above the noise level of the system, typically, three times the standard deviation of the blank ($n=8$). The limit of quantitation (LOQ) is the lowest level of analyte that can be accurately and precisely measured. The limit of quantitation, defined as 10 times the standard deviation of the blank ($n=8$). The limit of detection and limit of quantitation (LOQ) were of $(0.012464, 0.041546) \mu\text{g.ml}^{-1}$ respectively, depending on the standard deviation of the response and the slope of the calibration curve using the following equation: $(\text{LOD}=3\delta_B/s)$, $(\text{LOQ}=10\delta_B/s)$, where δ_B is the standard deviation of the blank calibration plot and (s) is its slope^(220,221). Comparing this with the traditional method, by direct aspiration of sample, the limit of detection and limit of quantitation where $(0.115111, 0.383704) \mu\text{g.ml}^{-1}$. The LOD and LOQ by using CPE method were 9 times smaller than the traditional. Figures (3.61) and (3.69) show these results. Furthermore, for preconcentration of 15ml of the working standard solutions based on the slope ratio of calibration curves with and without preconcentration⁽¹⁹¹⁾, an enrichment factor of 51.259, was obtained for Cd solution figure (3.70). To test the reproducibility of the proposed extraction method, the suggested procedure was repeated eight times under optimum conditions. The relative standard deviation (R.S.D.) was measured for $0.05\mu\text{g.ml}^{-1}$ to be 1.308%. The consumptive index is defined as the sample volume in milliliters consumed to reach a unit of enrichment factor (EF)⁽¹³²⁾: $\text{CI} = V_s(\text{ml}) / \text{EF} = 0.293$.

Table (3.15): Optimum conditions, performance characteristics for Cd-L₁

No.	Parameters	Value
1	Cd hollow cathode lamp element (nm)	228.80 nm
2	Sample volume (ml)	15ml
3	Chelating agent (L ₁) 5*10 ⁻³ M	1ml
4	HCl	0.5M
5	Incubation time (min)	20min
6	Water bath heating temperature(°C)	70°C
7	Triton X-100 (1%)	0.9ml
8	Slope with preconcentration	0.692
9	Slope without preconcentration	0.0135
10	Enrichment factor (Slope of CPE/Slope of Normal)	51.259
11	preconcentration factor	15
12	Consumptive index (CI)	0.293
13	Correlation coefficient (R ²)	0.9994
14	Concentration rang µg.ml ⁻¹	(0.01-1.3) µg.ml ⁻¹
15	limit of detection (3σB/slope) µg.ml ⁻¹	0.012464
16	Limit of quantitation (10δ/slope)µg.ml ⁻¹	0.041546
17	RSD% (n= 8) at 0.05µg.ml ⁻¹	1.308%



Figure(3.70): Enrichment factor between CPE method compare with normal determination

The previous figure shows the comparisons between preconcentration by CPE and the traditional measurements, there is a good enrichment of signal able to use it for detect micro quantity of cadmium in the sample .

3.9.7: Determination of cadmium ion in real samples

To evaluate the effectiveness of the developed procedure was applied for the determination of cadmium(II) in various samples including (milk, tobacco, tea, soil and two different water samples including river and tap water) in order to test its applicability and reliability. For this purpose, an aliquot of each sample prepared was preconcentrated following CPE recommended procedure (2.13), the results as shown in figure (3.71). Good recoveries (100.165-103.383%) were achieved for all analyzed samples by adding Cd(II) standard solution into the sample solution. FAAS was employed for the accuracy check, due to its better sensitivity. The results of this method for each type of the selected samples and the recoveries are shown in the table (3.16). As it can be seen, the determination of Cd²⁺ in real samples is noticeably accurate by the purposed method .

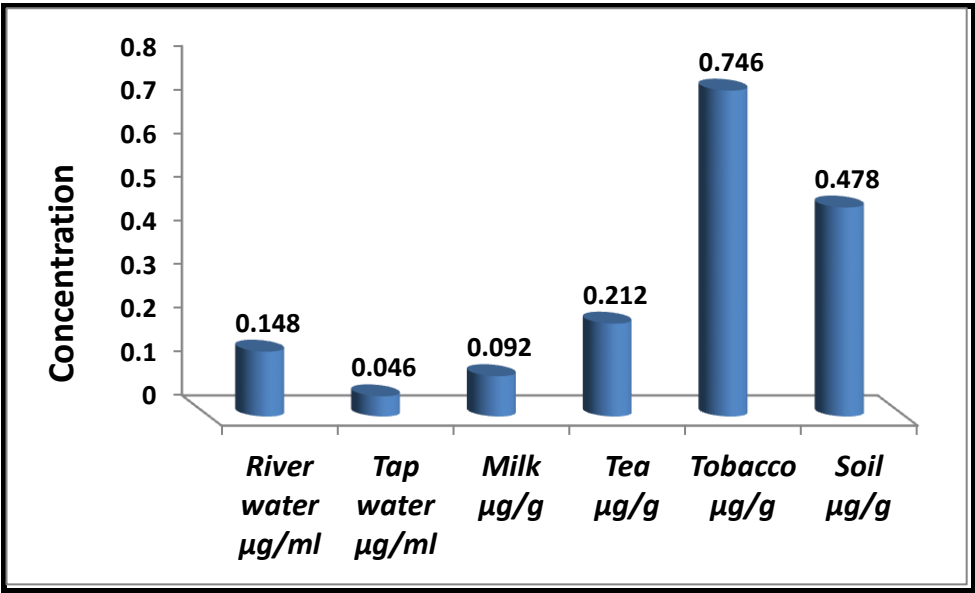


Figure (3.71): Determination of Cd^{2+} ion in real samples

Table (3.16): Recovery percentage of the cadmium ion real samples

Sample	Cd Add	Cd Found	% Recovery	%Error
River water ^a	0	0.148	-	-
	0.5	0.650	100.309	0.309
Tap Water ^a	0	0.046	-	-
	0.5	0.558	102.198	2.198
Milk ^b	0	0.092	-	-
	1	1.110	101. 648	1.648
Tea ^b	0	0.212	-	-
	1	1.214	100.165	0.165
Tobacco ^b	0	0.746	-	-
	1	1.766	101.145	1.145
Soil ^b	0	0.478	-	-
	1	1.528	103.383	3.383

a: All values are $\mu\text{g.ml}^{-1}$, b: All values are $\mu\text{g.g}^{-1}$

The results have shown that the concentration of Cd^{2+} ion in river and tap waters was just above the maximum permitted concentration ($0.003\mu\text{g}.\text{ml}^{-1}$) for protection of aquatic life and drinking water as reported by WHO(2011)⁽²²²⁾, while in milk too it is just more than the maximum permissible concentration ($0.01\mu\text{g}.\text{g}^{-1}$) as recommended by WHO⁽²²³⁾, The cadmium concentration in tea was found $0.212\mu\text{g}.\text{g}^{-1}$, which was above than the World Health Organization (WHO) recommended limit of $0.10\mu\text{g}.\text{g}^{-1}$ ⁽²²⁴⁾. Phosphate fertilizer applied to soil is a significant contributor for tea contamination for Cd ⁽²²⁵⁾. According to the MOA standard⁽²²⁶⁾, the Cd limit in tea was below from permissible concentration ($1\text{ mg}.\text{kg}^{-1}$).

Cadmium level in cigarette tobacco $0.746\mu\text{g}.\text{g}^{-1}$ was more in the literature range ($0.148\text{-}0.380\mu\text{g}.\text{g}^{-1}$)⁽²²⁷⁾ and above the permissible limit ($0.05\text{mg}/\text{kg}$)⁽²²⁸⁾. Cigarettes containing $0.5\text{--}3\mu\text{g}$ cadmium per gram of tobacco can result in up to $3\mu\text{g}$ daily cadmium absorption via the lungs, assuming a 25% absorption factor⁽²²⁹⁾. Tobacco plants have a special ability to absorb cadmium from soil. Concentration of Cd(II) in soil $0.478\mu\text{g}.\text{g}^{-1}$ was below the WHO/FAO⁽²³⁰⁾ permissible limit of $3\text{mg}/\text{kg}$ for soil. M.Ghaedi. *et.al* ⁽¹²¹⁾ was found concentration of cadmium in soil $0.370\mu\text{g}.\text{g}^{-1}$.

3.10: Cloud point extraction of copper (II)

3.10.1: Calibration curve of determination copper ion

A calibration curve was previously constructed for each experiments in this study in order to determine the quantity of Cu(II) ion. The absorbance was recorded in the concentration range of $(1\text{-}10)\mu\text{gml}^{-1}$ Cu(II) at λ_{max} of 324.75nm by flame atomic absorption spectrometry. The calibration curve was constructed by plotting values of absorbance versus concentration as shown in figure (3.72).

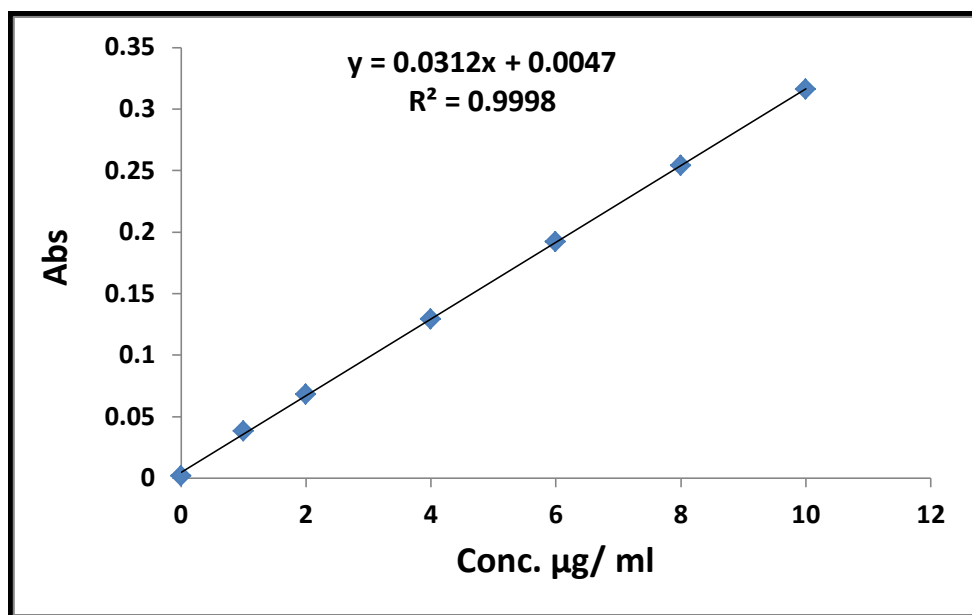


Figure (3-72): Calibration curve of copper(II) ion

3.10.2: Optimization of CPE procedure

The effects of several experimental parameters which impact the CPE efficiency were carried out by classical optimization (OFAT) procedure. In as much as the extraction efficiency of the CPE depends on dual factors, some of regarding the prior formation of a complex with sufficient hydrophobicity and the other for the formation of micelles to obtain the desired separation and preconcentration. Consequently, the effects of reagent concentration, HCl concentration, incubation time, equilibration temperature and Triton X-100 amount were selected in this study.

3.10.2.1: Effect of reagent (L_2) Concentration

The extraction according to CPE methodology depends on major step of complexation between organic reagent and Cu(II) which can be transferred to surfactant after formation cloud point phase. Since the complexation step also depends on the nature of organic reagent which specify the extraction capability via the formation more stable ion pair complex, the effect of reagent concentration was carried out by taking 10 ml solution containing $3\mu\text{g}.\text{ml}^{-1}$ Cu(II), 0.1 M HCl, 0.5ml of Triton X-100 plus various concentrations (1×10^{-4} , 5×10^{-4} , 1×10^{-3} , 5×10^{-3} , 1×10^{-2} , 5×10^{-2} M) of reagent (L_2). Each experiment was and subjected to the CPE process (2.13). The results are shown in figure(3.73). It was observed that the absorbance increased with increasing organic reagent concentration up to 5×10^{-3} M and suddenly absorbance decrease at higher concentration. It can be concluded that a concentration of 5×10^{-3} M of reagent gives more sensitivity and more stable ion pair complex extracted by CPE method.

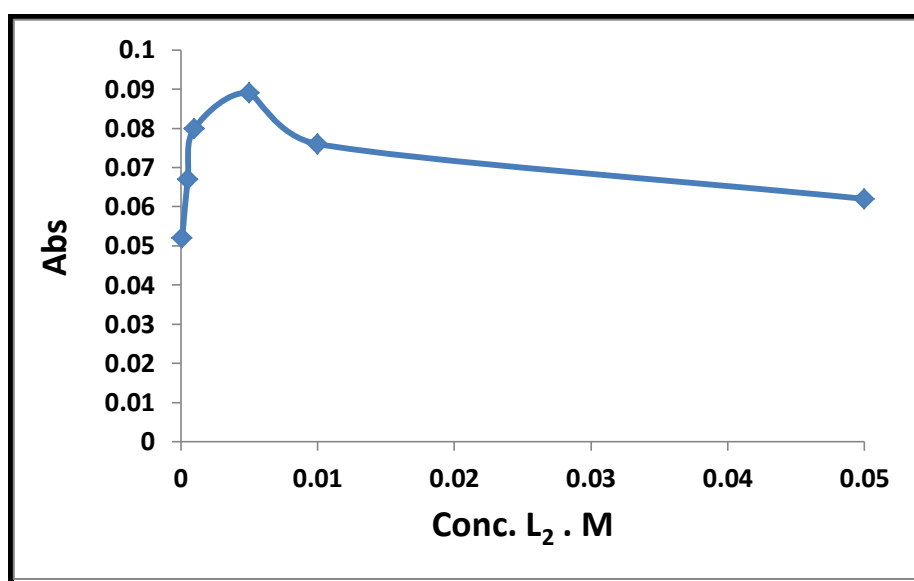


Figure (3.73): Effect L_2 concentration on absorbance

It can also be seen that the rate of formation of ion pair complex for extracted Cu(II) is faster than of back forward reaction at optimum concentration of organic reagent ($5 \times 10^{-3} \text{M}$), but at concentration less than optimum, the situation is different that the rate of back reaction becomes predominates leading to the dissociation of ion pair complex and decreased in absorbance⁽²¹⁹⁾.

3.10.2.2: Effect of HCl concentration on CPE

The separation Cu(II) ion by CPE method involves the formation of ion-association complex between this ion and the organic reagent(L_2) in the presence of HCl, to get sufficient hydrophobicity that can be extracted into micelle medium thus obtaining desired enrichment. The effect of HCl concentration was studied individually by measuring the absorbance of 10ml solution containing $3 \mu\text{gml}^{-1} \text{Cu(II)}$, $5 \times 10^{-3} \text{M}$ reagent solution (L_2) and 0.5ml of 1% Triton X-100 by varying the concentration range of (0.05-2)M HCl and each solution subjected to general CPE procedure (2.13). The results were depicted in the figure (3.74).

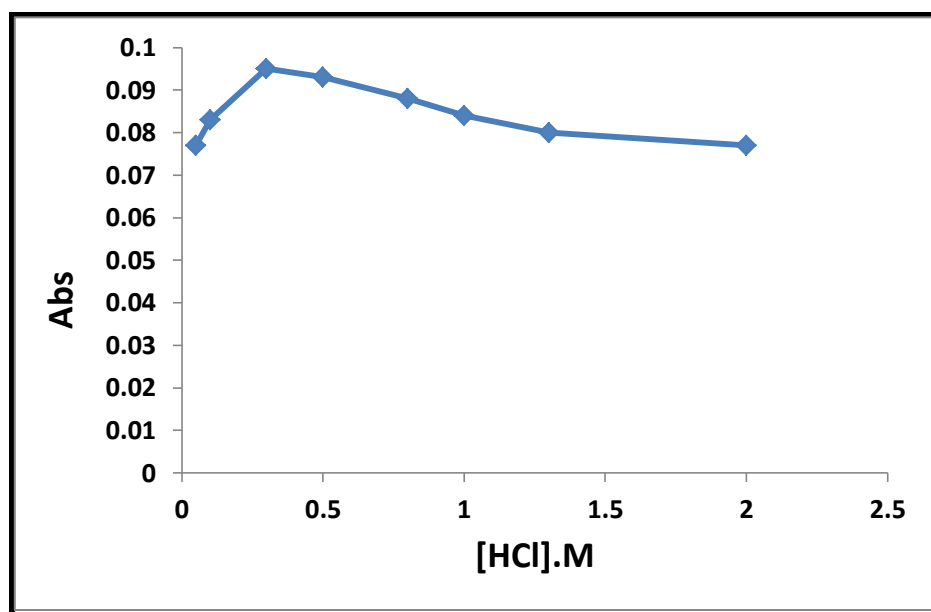
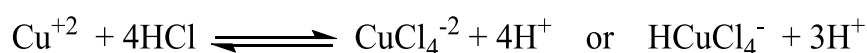
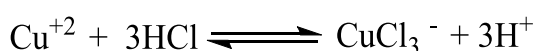
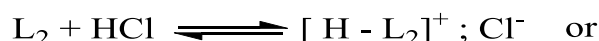
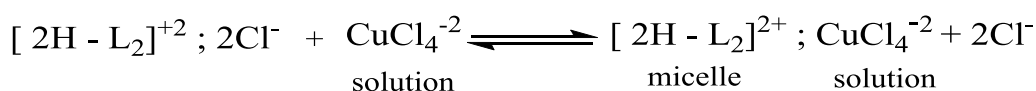
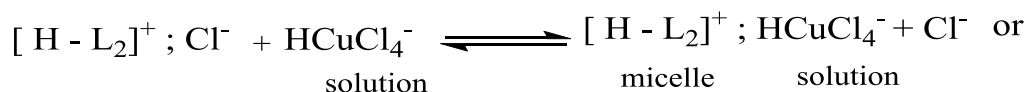


Figure (3.74): Effect of HCl concentration on absorbance

The results revealed that 0.3M of HCl was the optimum concentration, giving highest absorbance for the ion pair complex that extracted in micelles and more stable into surfactant-rich phase. Thus, the major effect of HCl was the formation of ion pair complex as shown below:



Thereafter, an exchange of small anions Cl^- with Cu ion may occur in micelles as the following :



3.10.2.3: Effect of Incubation Time

The incubation (heating) time is an important parameter in CPE process for a complete reaction to achieve an easy phase separation and the preconcentration to be as efficient as possible. The effect of time on the extraction of Cu(II) was conducted for the solutions containing 10 ml of $3\mu\text{gml}^{-1}$ of Cu(II) by varying the time between (10 - 45) min at 50°C and keeping other variables at optimal. It can be seen that the optimum heating time was 15 min for the extraction Cu(II) with L_2 . The results

have also shown that the absorbance increase with heating time to reach the optimal value (figures 3.75), indicating that better thermodynamic equilibria for cloud point formation with smaller volume, higher density of SRP, good dehydration and better partition of ion pair complex between two phases. From the kinetic point of view, this partition and dynamic movement and transference of ion pair complex to the formation cloud point layer were achieved at optimum time. At heating time of less than optimum value, no allowance for complete dehydration which might lead to increase in diffusion of micelles, thereby the extraction capability depressed. While at heating time of more than optimum value, the effect was also obvious in poorness of the extraction efficiency due to the increase of diffusion⁽²¹⁹⁾.

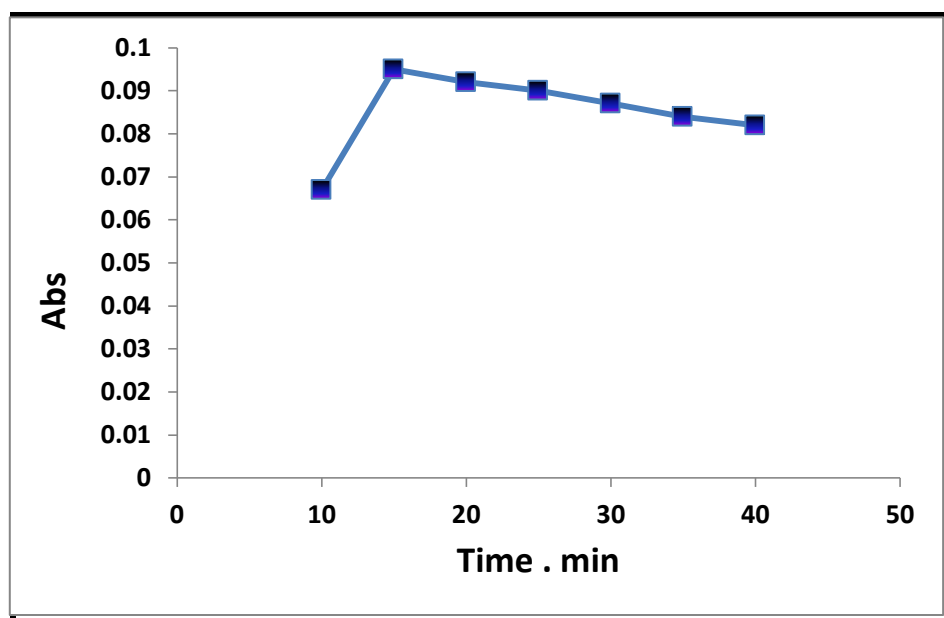


Figure (3.75): Effect of incubation time on absorbance

3.10.2.4: Effect of Equilibrium Temperature

The effect of temperature is also a crucial and very important parameter in CPE process to ensure phases separation and complete extraction in order to achieve preconcentration factor of an analyte efficiently as much as possible. Since extraction by CPE is performed under isothermal conditions for phase separation, an equilibration temperature that were well above the cloud point temperature of the surfactant is required to obtain the surfactant rich-phase with a small volume and a high density layer resulting from the micelles aggregation. The dependence of absorbance upon equilibration temperature was examined in the range of (30-90)°C at 15 min for the extraction of copper ion with (L₂), keeping other conditions at optimal. The results are shown in figure (3.76).

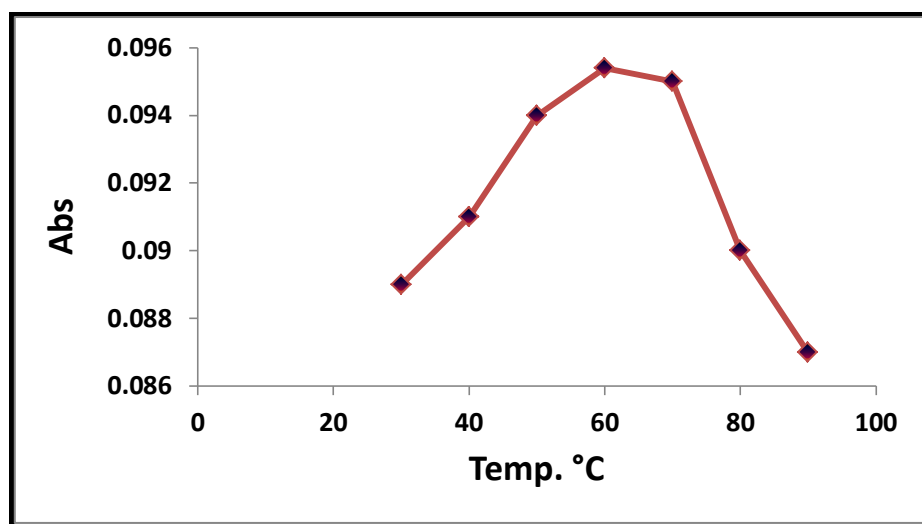


Figure (3.76) : Effect of heating temperature on absorbance

Preliminary experiments have indicated that at temperature below mbe formed due to a very low number of micelles formed leading to

difficulty in distinguishing the two phases thereby the ion-pair association complex which cannot be well separated. The results have shown that the optimum equilibrium temperature for extraction of copper ion with (L_2) was of 60°C at 15 min. This gave maximum absorption signals to be achieved, thus indicating a good quantitative extraction for Cu(II) ion. At a higher temperature than optimal, the absorption signal decrease due to the thermal decomposition of the complex⁽²¹⁹⁾. Therefore, 60°C at 15 min was selected for copper as the working equilibration temperature in this study.

3.10.3: Thermodynamic Study

It is useful to study the mechanism of phase separation by CPE which is still ambiguous and not intensively investigated yet. Therefore, in this study, some thermodynamic data is determinate for $H-L_2^+$; $HCuCl_4^-$ system using Triton X-100 as a mediated extracting agent in order to understand a probable mechanism above the cloud point temperature (CPT). Depending on different operation temperatures and the distribution of target species between two phases via calculating the distribution ratios (D), the equilibrium extraction constants (K_{ex}) can be calculated and subsequently thermodynamic parameters for the extraction of the complex system at various temperatures were also determined. The equilibrium constants (K_{ex}) at the selected temperatures were calculated from equation (2-2) mentioned in chapter two (2.14.4) and the results are presented in tables (3.17) and (3.18).

Table (3.17): Variation of temperature for the extraction copper by L_2 on distribution ratio D

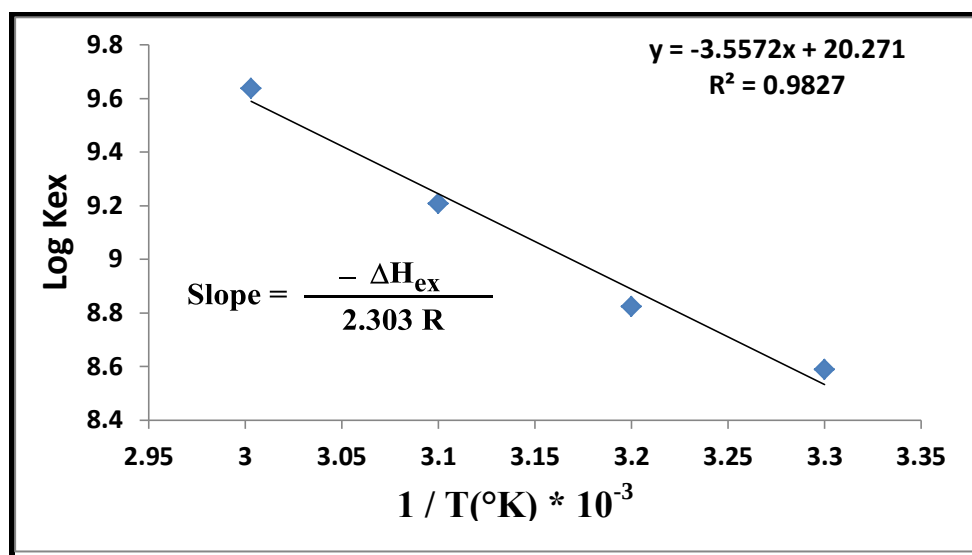
T / °C	T / °K	Absorbance	D
30	303	0.089	9.067
40	313	0.091	12.043
50	323	0.094	19.000
60	333	0.0954	31.468
70	343	0.095	27.571
80	353	0.090	10.152
90	363	0.087	7.499

Table (3.18): Variation of equilibrium constant with temperature during CPE for extraction Cu(II) with (L_2)

T°C	30	40	50	60
T°K	303	313	323	333
$1/T * 10^{-3}$	3.300	3.195	3.096	3.003
D	9.067	12.043	19.000	31.468
$K_{ex} * 10^9$	0.387	0.665	1.610	4.328
$\text{Log } K_{ex} * 10^9$	8.588	8.823	9.207	9.636

These thermodynamic equilibrium constants (K_{ex}) actually represent all equilibrium constants that affect the separation process such as aggregation micelles constant with increasing temperature, complex transference constant from aqueous phase to surfactant phase, association constant of the complex and distribution constant of the surface between aqueous phase and surfactant (micelles) phase. From the results in table (3-18), a plot of logarithm values of K_{ex} versus inverse temperature in Kelvin gave a straight line and from its slope, the enthalpy of extraction (ΔH_{ex}) can be calculated for CPE method as shown in the table (3.19). While the values of entropy (ΔS_{ex}) and Gibb free energy (ΔG_{ex}) of this extraction process are determined from the following thermodynamic

relationships [$\Delta G_{\text{ex}} = -RT \ln K_{\text{ex}}$] and [$\Delta G_{\text{ex}} = \Delta H_{\text{ex}} - T\Delta S_{\text{ex}}$] respectively that also described in chapter two (2.14.4). Once (ΔH_{ex}) and (ΔG_{ex}) are obtained, the ΔS_{ex} is calculated from second relationship. The results are presented in table (3.19). The values of enthalpy change (ΔH_{ex}) were obtained from the plot of $\log K_{\text{ex}}$ versus $1/T$ (figures 3.77) and found to be $0.0681 \text{ kJ mol}^{-1}$ for ion-pair complex of Cu, while the values of Gibbs free energy (ΔG_{ex}) and entropy change (ΔS_{ex}) were obtained at different temperatures from aforementioned thermodynamic relationships (2-2)-(2-6) in chapter two (2.14.4) as shown in table (3.19).



Figure(3.77): Extraction constant K_{ex} as a function of equilibrium temperature for ion pair complex of Cu^{2+} with(L_2)

Table (3.19): Thermodynamic parameters for the extraction of Cu ion-pair complex by CPE

$T/^{\circ}\text{K}$	ΔH_{ex} (KJ mol^{-1})	ΔG_{ex} (KJ mol^{-1})	ΔS_{ex} (J mol^{-1})
303	0.0681	-49.816	164.634
313		-52.869	169.128
323		-56.933	176.474
333		-61.434	184.691

It can be seen from table (3.19) that the enthalpy change (ΔH_{ex}) is equal to $0.0681 \text{ kJ mol}^{-1}$ indicating that reaction was endothermic, and the solubilisation process of $(\text{H-L}_2^+; \text{HCuCl}_4^-)$ complex is controlled by positive value of ΔH_{ex} which reflects efficiency of the extraction process of the complex that achieved thermodynamically into the surfactant-rich phase. This is explicit that a strong electrostatic association exists between Cu(II) anion with cationic reagent beside the contribution of the complex itself in driving water molecules out of surfactant phase in which more of micelles are aggregated enabling the precise extraction of complex especially in extracting of trace amounts. Accordingly, the extraction of ion-pair complex is easy and thermodynamically favourable and due to the positive value of ΔH_{ex} is achieved resulting in increasing the phase-volume ratio thus extraction efficiency.

On the basis of equation (2-5) mentioned in chapter two (2.14.4), ΔG_{ex} was found to be negative in all cases and its value increased approximately with temperature (table 3.19), indicating the extraction process is a spontaneous phenomenon because the complex transference and surfactant phase formation are synchronized processes that occur at The same time. Thus, the more negative value of ΔG_{ex} made the large spontaneous process. Accordingly, the spontaneity of ion pair complex extraction is governed by the negative value of $\Delta G_{\text{ex}}^{(217)}$, whilst the positive values of ΔS_{ex} prove that the solubilized ion-association complex molecules are organized in more random fashion during extraction process (i.e. entropic in region). Consequently, at optimum temperature (60°C), the micelles are quantitatively agglomerated with distinct lose of water so that they are accumulated in surfactant-rich phase and the

separation of phases occur due to the influence of internal energy of the micelles. Our findings were also revealed that from the value of enthalpy, the extraction forces of interdependence are electrostatics between copper anion and large cation of organic reagent degree of convergence between the two and therefore the interaction between the ion and organic reagent (L_2) is the interaction endothermic reaction. This is obvious where the values distribution ratio D increases with increasing temperature, which explains the increased correlation susceptibility between copper with L_2 increase with increasing temperature. This also confirms the possibility for strong association of organic reagent with ion metal, as well as the role of the reagent organic for increasing the coherence and removing the water molecules that surround metal ion^(216,219). The high values for entropy of extraction ΔS_{ex} show that the interaction is entropic in region and this enhances predominance balances of jump ion and the impact of organic reagent in helping to remove the water molecules from the anion and the increasing disorder irregular. It also shows that the effect of temperature and organic reagent molecules on the liberalization of the largest number of regular water molecules in the consistency of the casing metal anion and thereby forming the negative ion free (free ion) to increase the chances of contact and tighten the bonding between the organic reagent and the metal anion.

3.10.2.5: Effect of TX-100 Volume

It is known that the amount of surfactant used in CPE process for the separation for any analyte is a critical factor and its success depends on obtaining a maximum extraction efficiency and minimum phase volume ratio in order to increase the preconcentration factor. The effect of Triton X-100 amount was conducted by taking 10ml solution containing $3\mu\text{gml}^{-1}$ Cu(II), 5×10^{-3} (L_2), 0.3M HCl and varying volumes of (0.1-1.5) ml of 1% Triton X-100 in each case. The solutions were subjected to general CPE procedure (2.13). The results are seen in the figure (3.78).

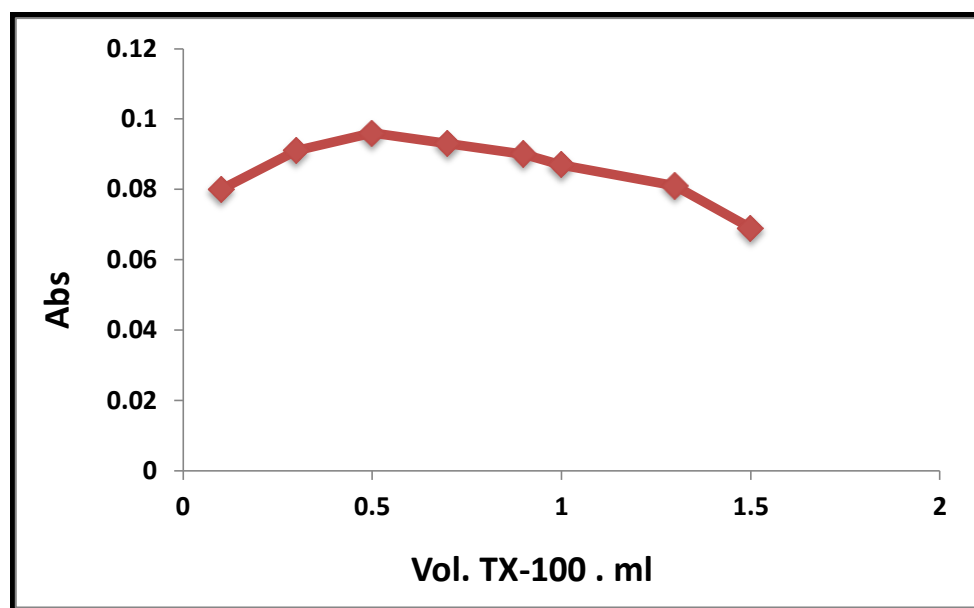


Figure (3.78): Effect volume of Triton X-100 on absorbance

It is shown that the maximal absorbance was achieved for ion-association complex extracted when the volume of 1% (w/v) Triton X-100 at 0.5ml for Cu(II) (figure3.78) and then the absorbance subsequently decreased sharply because of the increment in the overall analyte volumes and viscosity of surfactant-rich phase, or most probably due to

incomplete dehydration of cloud point layer may occur, thereby the two phases cannot be formed leading to poor sensitivity. On the other hand, at volume less than 0.5 ml of 1% Triton X-100 for Cu(II), a decrease in absorbance was observed due to insufficient extraction ion pair complex that formed⁽²¹⁹⁾.

3.10.4: Stoichiometry Determination of Complex (Slope Analysis Method)

It is reported that the analysis of the dependence $\log D = f(\log C_{L_2})$ permits the determination of the stoichiometry in the extracted complex in the table (3-20)⁽²²⁶⁾. Consequently, the slope in the figure (3.79) for $\log D = f(\log C_{L_2})$ coordinates is nearly equal to less than one, indicating the ion pair complexes with ratio 1:1 is extracted into the surfactant-rich phase. Thus, the most probable composition of the complex extracted of copper(II) as anion with organic reagent L_2 in the form of : $[H-L_2]^+; HCuCl_4^-$ or $[2H-L_2]^{2+}; CuCl_4^{2-}$.

Table (3.20): Effect of L_2 concentration on extractability of Cu(II)

$[L_2]$	Absorbance	D	Log D
1×10^{-4}	0.052	1.022	0.0095
5×10^{-4}	0.067	1.991	0.299
1×10^{-3}	0.080	4.190	0.622
5×10^{-3}	0.089	9.563	0.981

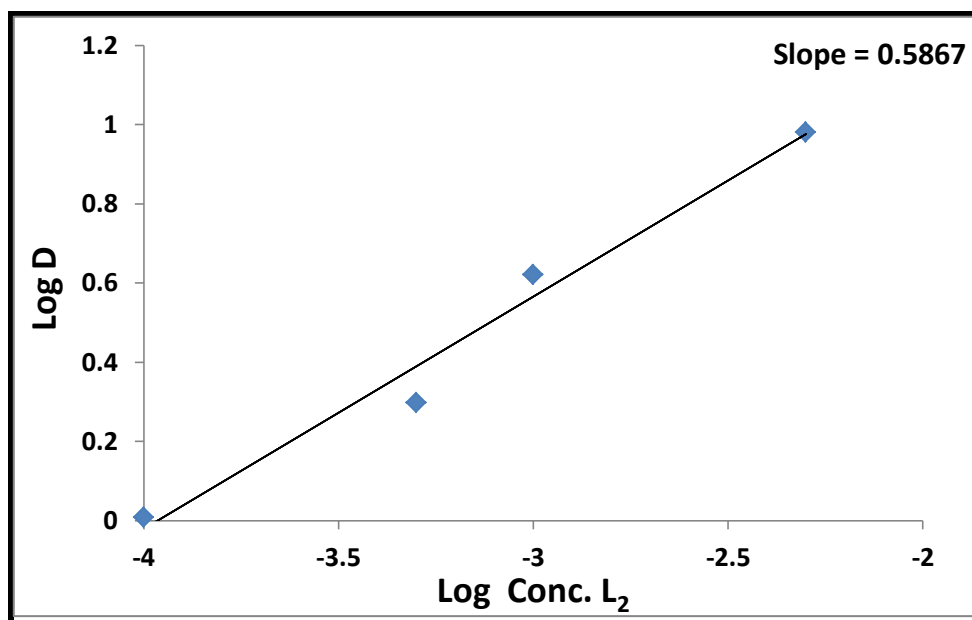


Figure (3-79): Slope analysis method: $\text{Log } D = f(\text{Log } C_{L_2})$

3.10.5: Calibration Curve of copper ion using CPE

The calibration graph is acquired from the optimized conditions like the data illustrated in figure (3.80). Calibration curve was obtained by preconcentrating of 15ml of standard solutions containing $(0.01-3)\mu\text{g.ml}^{-1}$ of Cu^{2+} under optimum conditions (concentration of the reagent (L_2), HCl, time, temperature and Triton X-100). The absorbance of final Solution was measured by FAAS. The correlation coefficient of the calibration curve equation was 0.9989 which indicates that a good linear regression was established between the absorbance and the concentration. The extracted solutions were introduced into the flame by conventional aspiration.

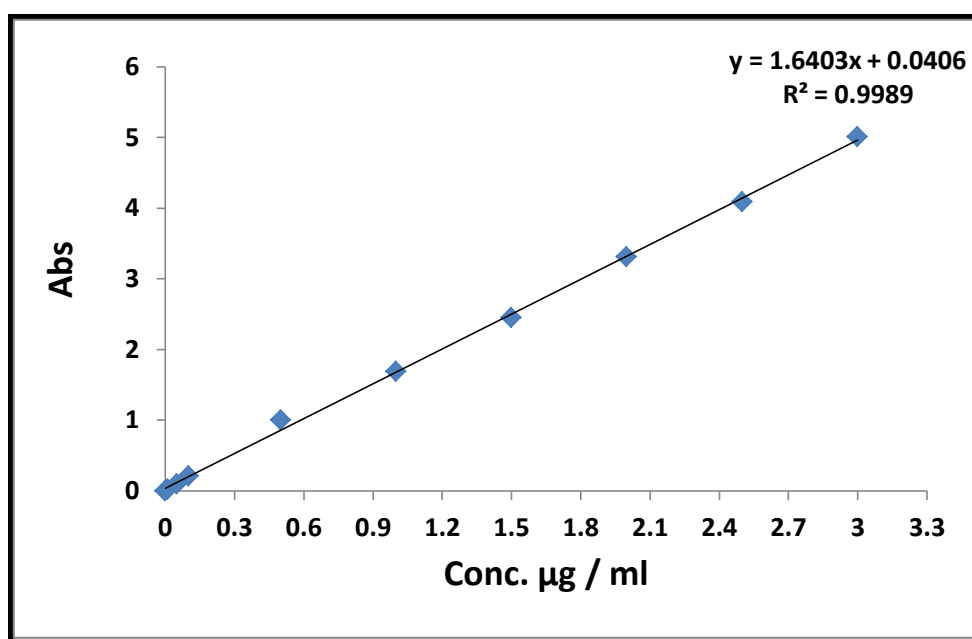


Figure (3.80): Calibration Curve for determination of copper ion using CPE-FAAS

The characteristics of the method was shown in table (3.21) give the calibration parameters for the purposed CPE method including the linear range, the relative standard deviation obtained for analyte sample subjected to the complete procedure, volume ratio and the limit of detection. Also, a calibration graph was obtained without preconcentration in order to calculate the enrichment factor. The enrichment factor was calculated as the ratio of the slopes of calibration curve with and without preconcentration .

3.10.6: Validation of Method and Calibration Curve of Copper Ion

Under the optimum conditions, performance characteristics were obtained by processing standard solutions of Cu^{2+} solution and the data is illustrated in table (3.21). Calibration functions were linear at least up to $(0.01 - 3\mu\text{g}.\text{ml}^{-1})$. The limit of detection (LOD) of a method is the lowest analyte concentration that produces a response detectable above the noise level of the system, typically, three times the standard deviation of the blank ($n=8$). The limit of quantitation (LOQ) is the lowest level of analyte that can be accurately and precisely measured. The limit of quantitation, defined as 10 times the standard deviation of the blank ($n=8$). The limit of detection and limit of quantitation (LOQ) were of $(0.017238, 0.057459)\mu\text{g}.\text{ml}^{-1}$, respectively. Comparing this with the traditional method, by direct aspiration of sample, the limit of detection and limit of quantitation were $(0.135211, 0.450641)\mu\text{g}.\text{ml}^{-1}$, respectively. The LOD and LOQ by using CPE method were 8 times smaller than the traditional. Figures (3.71) and (3.79) show these results. Furthermore, for preconcentration of 15ml of the working standard solutions based on the ratio of calibration curves with and without preconcentration⁽¹⁹¹⁾, an enrichment factor of 52.574, was obtained for Cu solution figure (3.81). To test the reproducibility of the proposed extraction method, the suggested procedure was repeated eight times under optimum conditions. The relative standard deviation (R.S.D.) without preconcentration for Cu was measured for $0.05\mu\text{g}.\text{ml}^{-1}$ to be 1.063%. The consumptive index is defined as the sample volume in milliliters consumed to reach a unit of enrichment factor (EF)⁽¹³²⁾: $\text{CI} = V_s(\text{ml}) / \text{EF} = 0.285$.

Table (3.21): Optimum conditions, performance characteristics for Cu-L₂

No.	Parameters	Value
1	Cu hallow cathode lamp element (nm)	324.75nm
2	Sample volume (ml)	15ml
3	Chelating agent (L ₂) 5*10 ⁻³ M	1ml
4	HCl	0.3M
5	Triton X-100 (1%)	0.5ml
6	Water bath heating temperature(°C)	60°C
7	Incubation time (min)	15min
8	Slope Without preconcentration	0.0312
9	Slope with preconcentration	1.6403
10	Enrichment factor (Slope of CPE/Slope of Normal)	52.574
11	preconcentration factor	15
12	Correlation coefficient (R ²)	0.9989
13	Concentration rang µg.ml ⁻¹	(0.01 - 3) µg.ml ⁻¹
14	Limit of detection (3σ/slope) µg.ml ⁻¹	0.017238
15	Limit of quantitation (10σ/slope) µg.ml ⁻¹	0.057459
16	Consumptive index (CI)	0.285
17	RSD% (n= 8) at 0.05µgml ⁻¹	1.063%

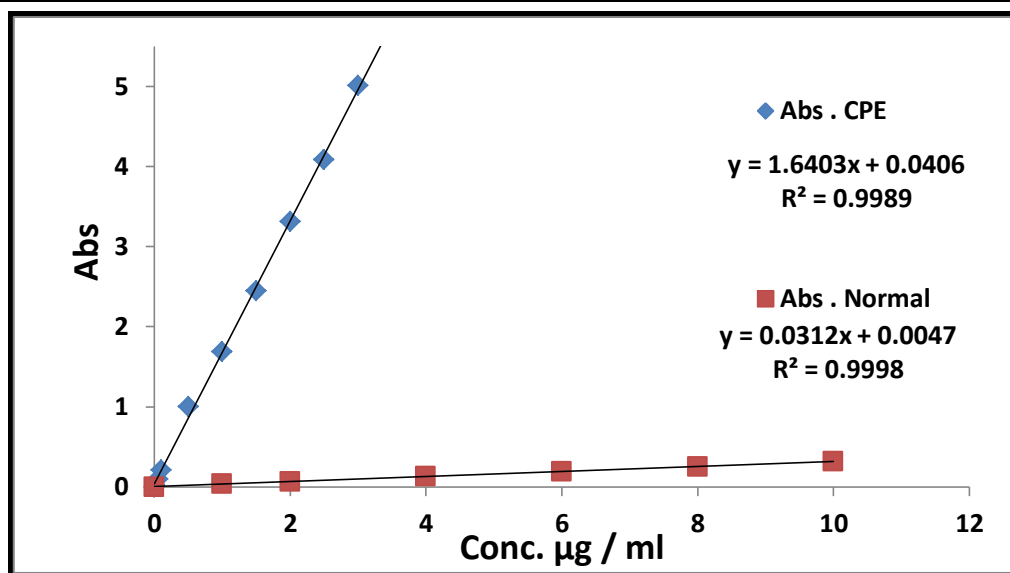


Figure (3.81): Enrichment factor between CPE method compare with normal determination

The previous figure shows the comparisons between preconcentration by CPE and the traditional measurements, there is a good enrichment of signal able to use it for detecting micro quantity of copper in the sample .

3.10.7: Determination of copper ion in different real samples

To evaluate the effectiveness of the developed procedure for the determination of Cu^{2+} ion in real samples including (milk, tobacco, tea and two different water samples including river and tap water) were applied the results as shown in figure (3.82). Good recoveries (100.380-103.377%) were achieved for all analyzed samples by adding Cu(II) standard solution into the sample solution FAAS which was employed for the accuracy check, due to its better sensitivity. The results of this method for each type of the selected samples and the analytical recoveries are shown in the table (3.22). As it can be seen, the determination of Cu^{2+} ion in real samples is noticeably accurate by the purposed method.

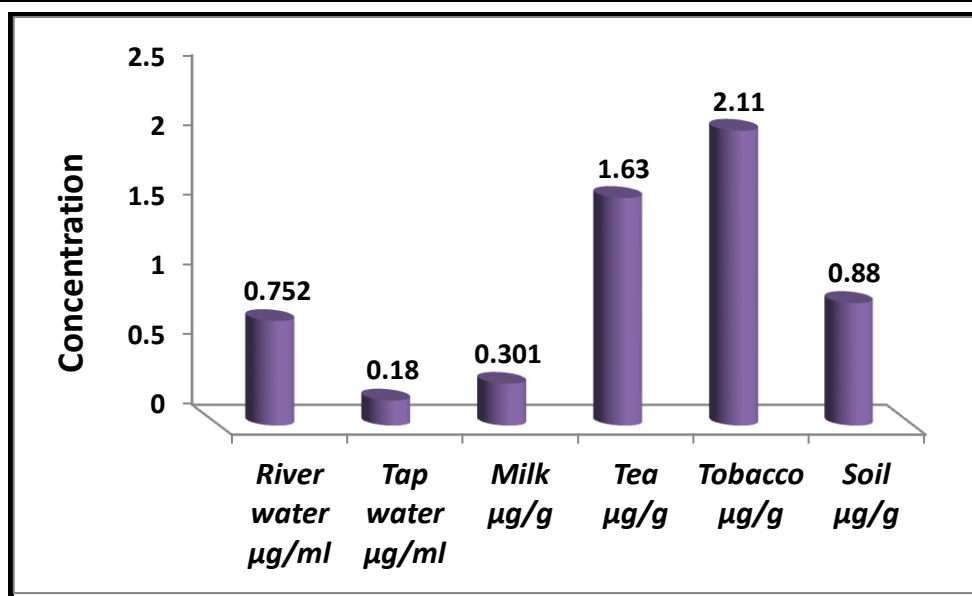
Figure (3.82): Determination of Cu²⁺ ion in real samples

Table (3.22) : Recovery percentage of the copper ion real samples

Sample	Cu Add	Cu Found	% Recovery	%Error
River water ^a	0	0.752	-	-
	0.5	1.268	101.278	1.278
Tap Water ^a	0	0.180	-	-
	0.5	0.684	100.588	0.588
Milk ^b	0	0.301	-	-
	1	1.350	103.377	3.377
Tea ^b	0	1.630	-	-
	1	2.640	100.380	0.380
Tobacco ^b	0	2.110	-	-
	1	3.131	100.675	0.675
Soil ^b	0	0.880	-	-
	1	1.940	103.191	3.191

a: All values are µg.ml⁻¹, b: All values are µg.g⁻¹

The results have shown that the concentration of Cu^{2+} in river and tap water was less the maximum permitted concentration ($2\mu\text{g.ml}^{-1}$) in water according to WHO^(118,232). The maximum limit concentration of copper in milk according to WHO and Joint Expert Committee on food and Agriculture is 0.01mg/l ⁽²³³⁾, in the present study was just more than from limit but less than the maximum permissible concentration (0.4mgkg^{-1}) as recommended by (EC,2001)⁽²³⁴⁾. The copper concentration in tea was found $1.630\mu\text{g.g}^{-1}$, the maximal content of Cu^{2+} in tea is limited in some regulations. According to the regulations imposed upon tea in Japan (100 mg/kg) and the United States (150mg/kg)⁽²³⁵⁾, according to the Industrial Standard (NY/T288-2012) of China's Ministry of Agriculture (MOA)⁽²³⁶⁾, the Cu^{2+} content was limited to 30 mg/kg . The determined levels of Cu^{2+} in the tea sample was below from all the limits. Copper level in cigarette tobacco was $2.110\mu\text{g.g}^{-1}$ below the permissible limit by WHO (10 mg/kg)⁽²²⁸⁾. The concentration of copper recorded in soil was below the WHO/FAO (2001) permissible limit of 100 mg/kg for soils⁽²³⁰⁾, but the copper content in soil above in the literature⁽²³⁷⁾ ($0.430\mu\text{g.g}^{-1}$).

3.11: Cloud-Point Extraction and Determination of Cobalt ion

A new and versatile procedure for the determination of cobalt using micelle-mediated extraction is developed. The method which involves the formation of an ion association complex between cobalt and reagent (L_3) in acidic solution and the complex is extracted into the surfactant Triton X-100 at optimum conditions. The surfactant-rich phase which contains cobalt complex is mediated with solvent and the Co^{2+} content measured FAAS at absorption maximum λ_{max} of 240.72 nm. The effect of the several variables which impact the CPE efficiency are optimized by one-factor-at-a-time(OFAT). The developed method was used for the determination of cobalt in river water, tap water, tea, milk, tobacco and soil samples.

3.11.1: Calibration curve of determination cobalt ion

A calibration curve was previously constructed for each experiments in this study in order to determine the quantity of Co(II) ion. The absorbance was recorded in the concentration range of $(0.1-8)\mu g.ml^{-1}$ Co(II) at λ_{max} of 240.72nm by flame atomic absorption spectrophotometer (FAAS). The calibration curve was constructed by plotting values of absorbance versus concentration as shown in figure (3.83) .

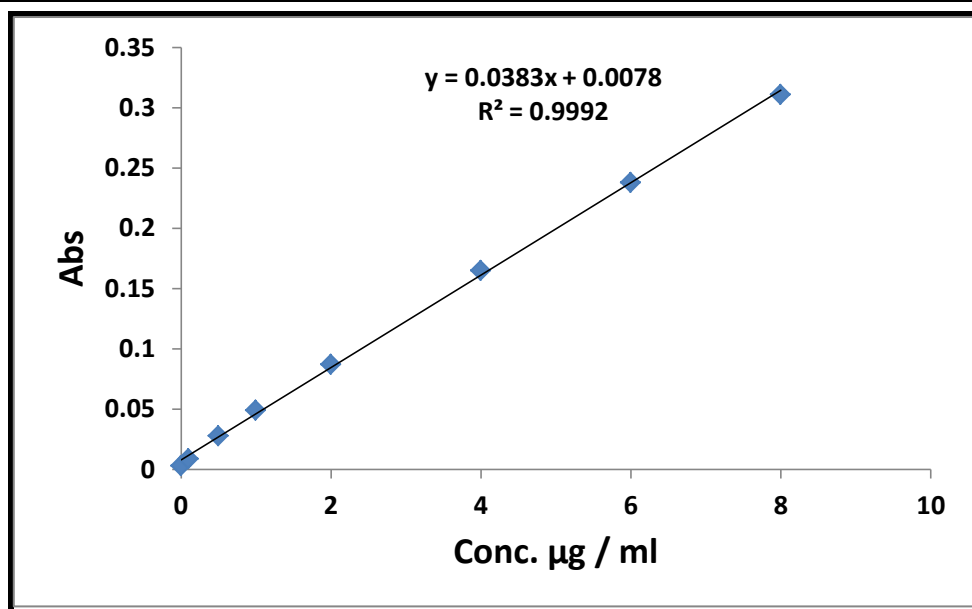


Figure (3.83): Calibration curve of cobalt (II) ion

3.11.2: Optimization of CPE Procedure

The effects of several experimental parameters which impact the CPE efficiency were carried out by classical optimization (one-variable-at-a-time). In this approach, we observe the effect of one factor at a time (OVAT) on an experimental response. While only one factor is changed, others are kept at a constant level. Although, the “optimization” performed by OVAT⁽²¹²⁾ does not ensure at all that the real optimum will be conformed, but it would be valid only if the variables to be optimized would be totally independent from each other (i.e. no interactive effects among the variables). Nevertheless, the classical optimization certainly leads at least to an improvement of the analytical method. Inasmuch as the extraction efficiency of the CPE depends on dual factors, some of regarding the prior formation of a complex with sufficient hydrophobicity and the other for the formation of micelles to obtain the desired

separation and preconcentration. Consequently, the effects of ligand concentration, HCl concentration, incubation time, equilibration temperature, Triton X-100 volume were selected in this study.

3.11.2.1: Effect of reagent (L_3) Concentration

The variation of absorption signal as a function of reagent concentration was investigated for 10 ml solution containing $3\mu\text{g ml}^{-1}$ Co(II), 0.5ml of 1% Triton X-100, 0.1M HCl and varying concentrations of (1×10^{-4} , 5×10^{-4} , 1×10^{-3} , 5×10^{-3} , 1×10^{-2} , 5×10^{-2} M) of reagent(L_3) followed CPE procedure (2.13). The results are presented in the table (3.23), figures (3.84) and (3.85). For the determine remainder quantity of Co(II) in aqueous solution after extraction by subtraction quantity of Co(II) measured in the surfactant-rich phase from original quantity, afterward calculate Distribution ratio (D).

The results have shown that the analytical response increased with increasing in the concentration of L_3 reagent and reached maximum up to 1×10^{-3} M (L_3), indicating sufficient for ion-pair complex formation. Thus a concentration of 1×10^{-3} M of L_3 was selected as optimum. The results also revealed that the concentration of 1×10^{-3} M of L_3 provides a high distribution ratio (D) with highly stable ion pair complex formation extractable into surfactant (figure 3.84). Since CPE extraction procedure is in thermodynamic behaviour of extracted species^(238,239), It might be divided into two steps: the first step is the formation of complex between L_3 and Co^{2+} and the second step is the extraction of this complex into the cloud point phase. Thus, L_3 concentration plays effective role for the formation of ion pair complex according to the thermodynamic equilibria.

When the concentration of L_3 less than $1 \times 10^{-3} M$, it is not sufficient to reach optimum thermodynamic equilibria for complex formation because the rate of dissociation became more than rate of association if the concentration of L_3 is more than $1 \times 10^{-3} M$.

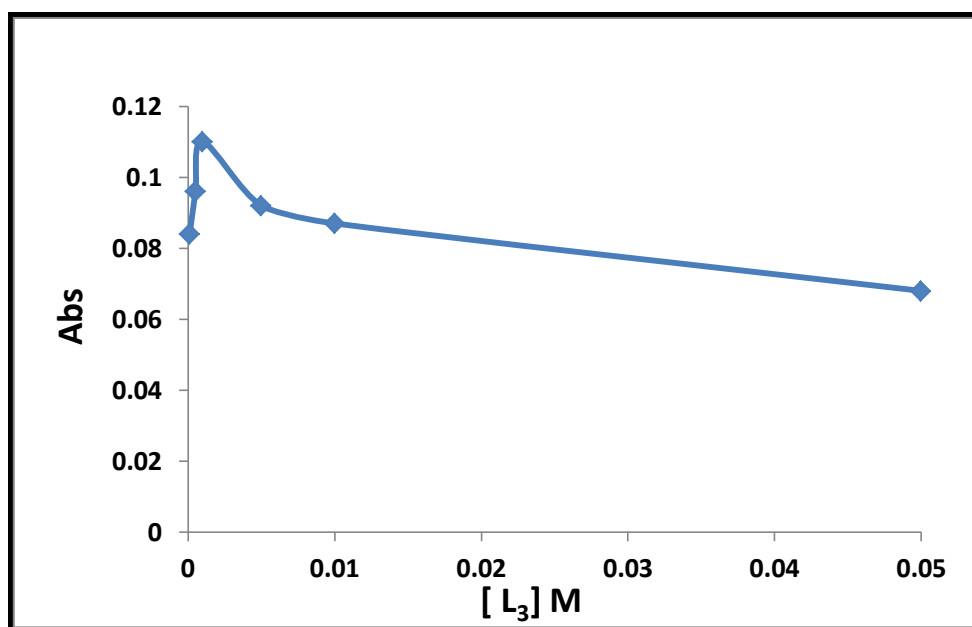


Figure (3.84): Effect L_3 concentration on the absorbance

Table (3.23): Effect of L_3 concentration on absorbance and distribution ratio (D)

$[L_3]$	Absorbance	D	Log D
1×10^{-4}	0.084	1.938	0.287
5×10^{-4}	0.096	3.243	0.511
1×10^{-3}	0.110	8.063	0.906
5×10^{-3}	0.092	2.686	0.429
1×10^{-2}	0.087	2.202	0.343
5×10^{-2}	0.068	1.112	0.046

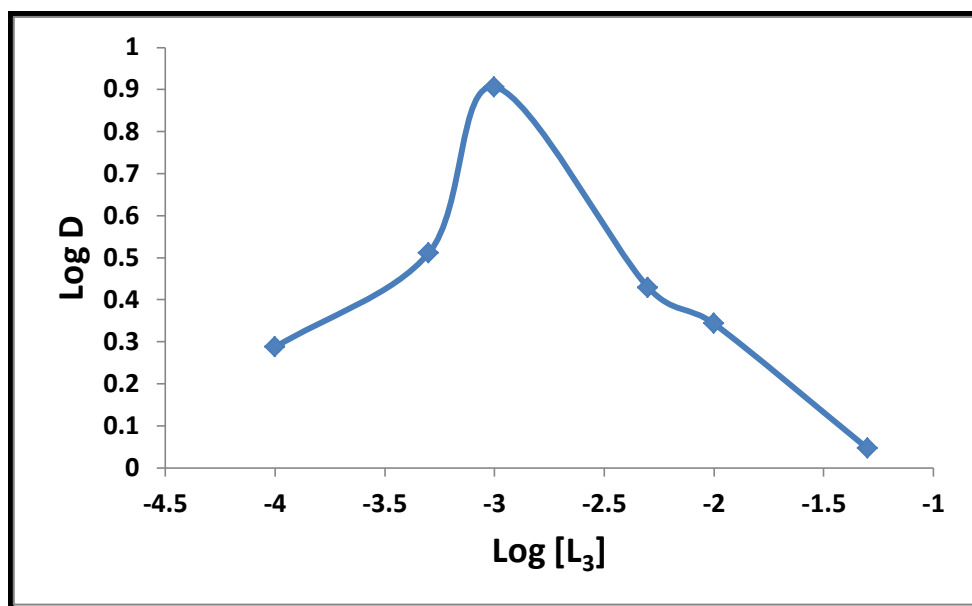


Figure (3.85): Effect of L_3 concentration on distribution ratio

3.11.2.2: Effect of HCl Concentration

The separation of Co(II) by CPE method involves the formation of ion-association complex between Co(II) and the protonated reagent in the presence of HCl, with sufficient hydrophobicity and extracted into micelle medium thus obtaining desired enrichment. In this study, 10ml solution containing $3\mu\text{g.ml}^{-1}$ Co(II), reagent (L_3) 1×10^{-3} M and 0.5 ml of 1% Triton X-100 and varying concentration range of (0.05-2) M HCl was subjected to general CPE procedure (2.13). The results in figure (3.86) show the effect of HCl concentration on the absorbance and extractability of ion-association complex into the surfactant-rich phase. The results illustrate that the concentration at 0.8M HCl was found to be optimum for extraction ability according to CPE method. The absorbance increases with increase HCl concentration and reaching maximum at 0.8M HCl, referring to an adequate extraction efficiency of $[\text{CoCl}_4^{2-}][L_3^+]$ ion-pair in

water - nonionic surfactant phase system (figure 3.86). At less than 0.8M HCl, no thermodynamic equilibria can be reached for extraction of ion pair complex in micelle cloud point, thus the probability of complex formation might not be achieved of which a decrease in absorbance and distribution ratio D values, while at higher than 0.8M HCl, no suitable extraction may be occurred due to the deviation of thermodynamic equilibria which leads to dissociate the ion-pair complex according to mass action law and Le Chatlier principle and hence the absorbance suddenly decreases (figure 3.86) giving lower extraction efficiency⁽²³⁸⁾.

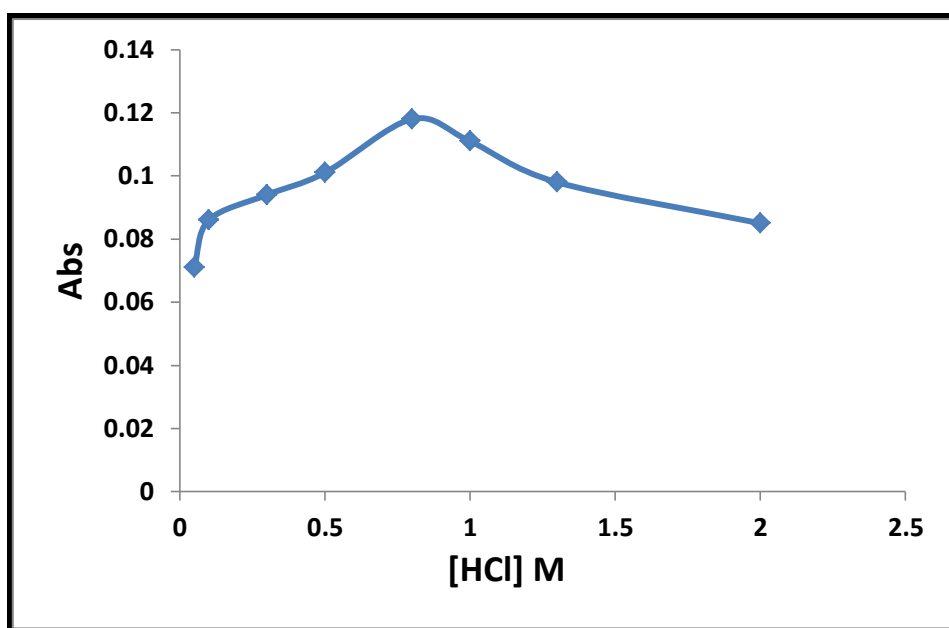


Figure (3.86): Effect of HCl concentration on absorbance

3.11.2.3: Effect incubation time

The effect incubation time on extraction is an important parameter in CPE process for a complete reaction to achieve easy phase separation and the preconcentration to be as efficient as possible. The effect of time on the extraction of Co(II) in the range of (10-45) min was investigated by taking 10ml aqueous solution containing $3\mu\text{gml}^{-1}$ Co^{2+} , 0.8M HCl, $1 \times 10^{-3}\text{M}$ of (L_3), 0.5ml of 1% (w/v) Triton X-100, followed CPE procedure (2.13). The results are shown in figure (3.87).

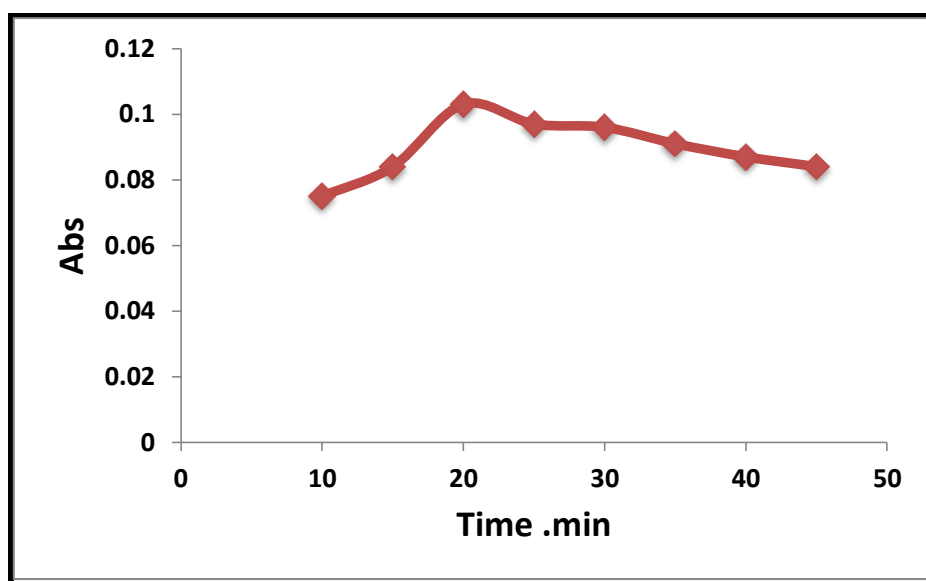


Figure (3.87): Effect of time on absorbance

It was also observed that the incubation time of 20min was sufficient for the maximum absorbance of cobalt complex (figure 3.87). This optimum time is most valuable time to formation CMC with high density and reach a suitable kinetic energy which give a good aggregation for surfactant in obtaining a smaller volume and the highest of a cloud point layer. Whereas, time of less than 20 min cannot allow to reach

maximum kinetic equilibrium for for aggregation micelles to form CMC and decline extraction efficiency. But at time of heating of more than 20 min, effect of increasing the diffusion of micelles in aqueous solution by increasing the kinetic energy is more than needed. Thus a decrease in density and increase in volume of cloud point layer occur resulting in minimizing extraction efficiency also⁽²³⁹⁾.

3.11.2.4: Effect of Equilibrium Temperature

To ensure phase separation and preconcentration of an analyte efficiently, optimal equilibration temperature is very crucial parameter for complete reaction which reflect certainly the magnitude of preconcentration factor of an analyte. The effect of the equilibrium temperature was examined by taking 10ml aqueous phase containing of $3\mu\text{g.ml}^{-1}$ Co(II) and keeping other factors at optimum conditions. The temperature was varied from 30 to 90°C at 20 min in a search of optimum value. It was shown that the highest absorbance signal was obtained when the temperature at 60°C achieving quantitative extraction (figure 3.88). Certainly, high temperatures are not suitable for the CPE procedure because these could cause problems of the stability of complex due to thermal decomposition of ion-association complex. In this study, 60°C was chosen as the optimized incubation temperature for all experiments.

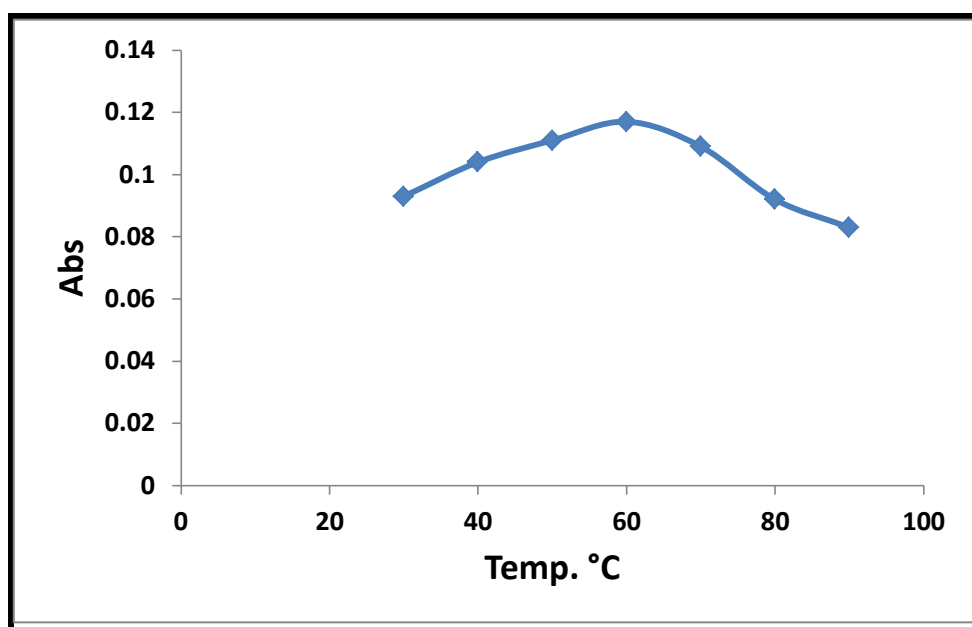


Figure (3.88): Effect of temperature on absorbance

3.11.3 : Thermodynamic Study

The equilibrium extraction constants (K_{ex}) and thermodynamic parameters for the extraction of $[L_3^+ ; CoCl_4^{2-}]$ complex during the cloud point extraction using Triton X-100 as a mediated extracting agent at temperature were determined 60°C. The equilibrium constants (K_{ex}) at the selected temperature were calculated from the equation mentioned in (2.14.4) in chapter two and the results are shown in figure (3.89). These thermodynamic equilibrium constants (K_{ex}) actually represent all equilibrium constants that affect the separation process such as aggregation micelles constant with increasing temperature, complex transference constant from aqueous phase to surfactant rich-phase, association constant of the complex and distribution constant of the surface between aqueous phase and surfactant (micelles) phase. The results are summarized in table (3.24) to (3.26). The value of enthalpy

change (ΔH_{ex}) was obtained from the plot of $\log K_{\text{ex}}$ versus $1/T$ (figure 3.89) and found to be $0.099 \text{ kJ mol}^{-1}$ while the values of Gibb's free energy (ΔG_{ex}) and entropy change (ΔS_{ex}) were obtained at different temperatures from thermodynamic relationships. Thermodynamic parameters were also calculated from the relationships mentioned in (2.14.4).

Table (3.24): Variation of temperature for the extraction cadmium by L_3 on distribution ratio D

T / °C	T / °K	Absorbance	D
30	303	0.093	2.861
40	313	0.104	5.036
50	323	0.111	9.417
60	333	0.117	19.408
70	343	0.109	7.721
80	353	0.092	2.722
90	363	0.083	1.863

Table (3.25): Variation of equilibrium constant with temperature during CPE for extraction Co(II) with (L_3)

T°C	30	40	50	60
T°K	303	313	323	333
$1/T * 10^{-3}$	3.300	3.195	3.096	3.003
D	2.861	5.036	9.417	19.408
$K_{\text{ex}} * 10^8$	2.170	5.972	19.269	77.819
$\log K_{\text{ex}} * 10^8$	8.336	8.776	9.285	9.891

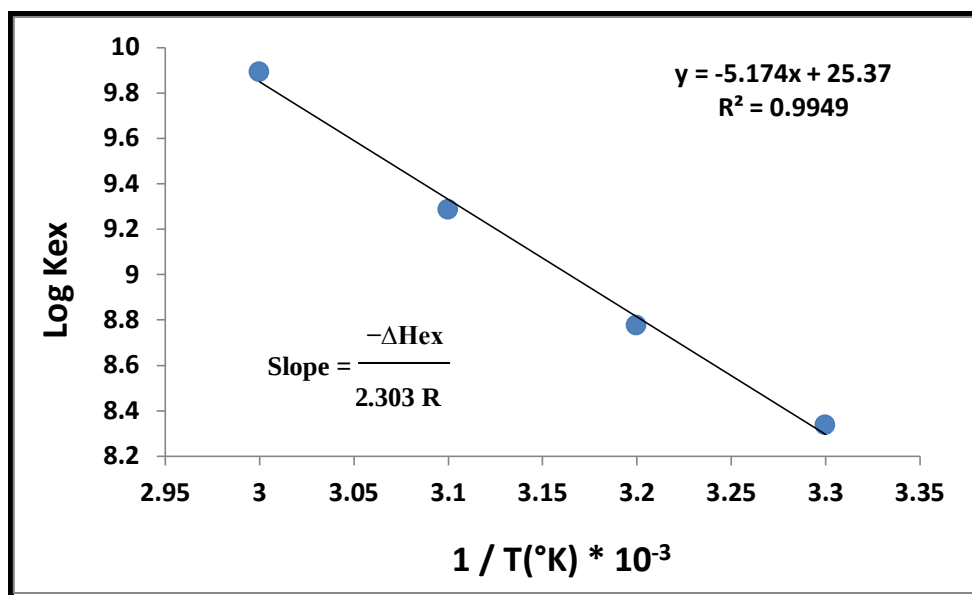


Figure (3.89): Effect of the equilibration extraction temperature on ion pair complex formation by CPE

It can be seen from table (3.26) that the enthalpy change (ΔH_{ex}) is equal to $0.099 \text{ KJ mol}^{-1}$ indicating a high efficiency of the extraction process of complex was achieved thermodynamically into the surfactant-rich phase. This is because (1) a strong electrostatic association exists between CoCl_4^{2-} anion with the reagent cation L_3^+ and (2) contribution of the complex in driving water molecules out of surfactant phase in which more of micelles are aggregated enabling the precise extraction of complex especially in extracting of trace amounts. As for ΔG_{ex} is negative in all cases, indicating the extraction process is a spontaneous phenomenon because the complex transference and surfactant phase formation are synchronized processes that occur at the same time. Thus, the more negative value of ΔG_{ex} , the large spontaneous process. The positive values of ΔS_{ex} prove that the solubilized ion-association complex molecules are organized in more random fashion during extraction

process (i.e. entropic in region). Therefore, the extraction process is endothermic in nature, which is also verified from the positive value of ΔH_{ex} (table 3.26).

Table (3.26): Thermodynamic parameters for the extraction of Co ion - pair complex by CPE

T°K	ΔH_{ex} (KJ mol ⁻¹)	ΔG_{ex} (KJ mol ⁻¹)	ΔS_{ex} (J mol ⁻¹)
303	0.099	-48.359	159.927
313		-52.589	168.332
323		-57.415	178.062
333		-63.058	189.661

The result (table 3.26) appear extraction method of Co²⁺ ion according to CPE methodology was endothermic and the low value of ΔH_{ex} reflect the high approach of ions in ion pair association complex extracted and high negative value of ΔG_{ex} illustrate the spontaneously of extraction method as well as high the positive value of ΔS_{ex} mean the extraction method depend on the change from systematic to unsystematic by dehydration and destroy the hydration shell of ions as well as change in the hydrogen bonds lattice and called this method is (entropic region)⁽²³⁸⁾.

3.11.2.5 : Effect of TX-100 Volume

A successful cloud point extraction procedure should maximize the extraction efficiency by minimizing the phase volume ratio, which improves its enrichment ability. The effect of the surfactant amount on CPE of 10 ml containing $3\mu\text{g}\cdot\text{ml}^{-1}$ Co(II) with 1×10^{-3} M L_3 at 0.8 M HCl in the presence of 1% Triton X-100 varying from (0.1-1.5) ml followed the general CPE procedure (2.13) has been optimized in order to obtain maximum extraction efficiency via minimizing the phase volume ratio. The results are shown in figure (3.90) .

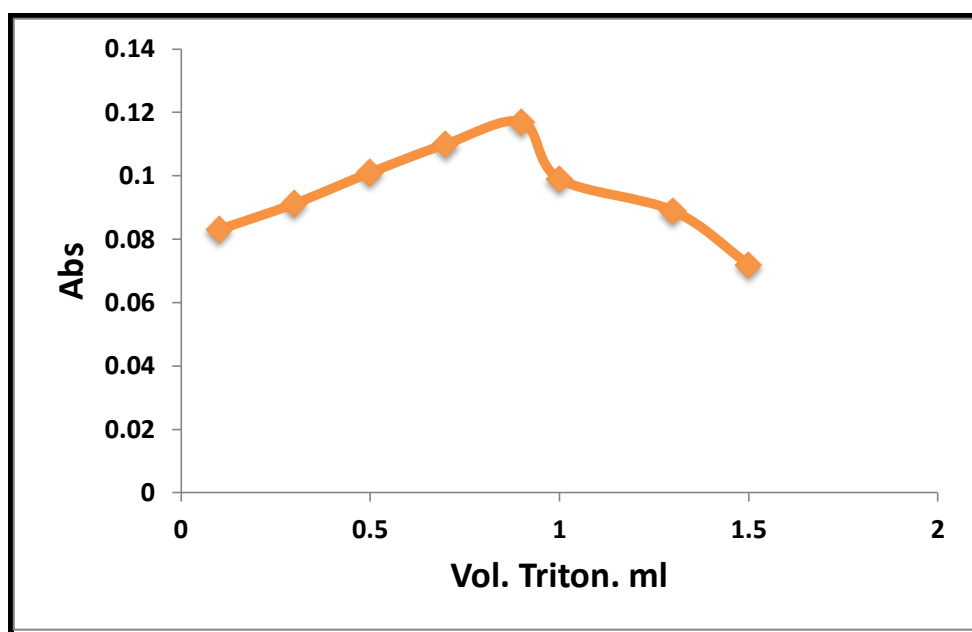


Figure (3.90): Effect of Triton X-100 amount on absorbance

Thermodynamic equilibrium for extraction and preconcentration for any analyte needs a significant amount of surfactant in order to form cloud point phase layer with higher viscous and smaller volume available for

the extraction of a major quantity of ion pair complex by giving higher absorbance and D value. Figure (3.88) reflects the impact of Triton X-100 amount on the preconcentration of Co(II). It was shown that the absorbance signal increases dramatically and reaches the maximum at 0.9 ml of 1% Triton X-100 and decreases suddenly. At a low amount of surfactant, the absorbance for ion pair complex are low, perhaps due to the insufficiency of assemblies formation entrapping the complex quantitatively. Whilst at a higher amount of surfactant, the extraction efficiency is low, probably due to the increase in surfactant-rich phase volume at which the analyte become more diluted (i.e. not good aggregation by effect of diffusion in aqueous phase) or according to thermodynamic behaviour of Triton X-100 which cannot allow to form smaller volume and higher viscous layer resulting in lowest extraction. Consequently, optimum volume of 1% (w/v) Triton X-100 at 0.9 ml was gave a high sensitivity and the highest extraction efficiency for the ion-pair complex.

3.11.4: Stoichiometric Determination of Complex

(Slope Analysis Method)

The stoichiometric determination for the reaction of CoCl_4^{2-} anion with L_3^+ organic reagent forming an ion-association complex was obtained according to the procedure mentioned in (2.16) by the dependence of $\log D = f(\log C_{\text{L}_3})$. The results are previously presented in table (3.23) and shown in figure (3.91) .

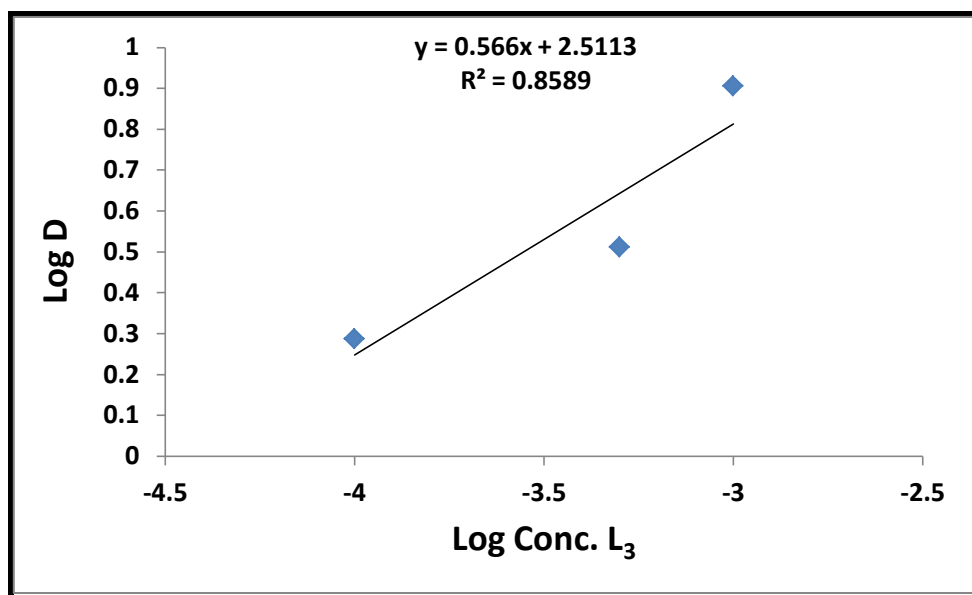


Figure (3-91): Slope analysis method: $\log D = f(\log C_{L3})$

The results revealed that the slope of the figure (3.91) in $\log D = f(\log C_{L3})$ coordinates is equal to 0.566, indicating the ion pair complex with Co: L_3 ratio of 1:1 is extracted into the surfactant-rich phase.

3.11.5: Calibration Curve of cobalt ion using CPE

The calibration graph is acquired from the optimized condition like the data illustrated in figure (3.92). Calibration curve was obtained by preconcentrating of 15ml of standard solutions containing (0.025-3 $\mu\text{g. ml}^{-1}$) under the optimum conditions (concentration of the reagent (L_3), HCl, incubation time, temperature and Triton X-100). The absorbance of final solution was measured by FAAS. The correlation coefficient of the calibration curve equation was 0.9966 which indicates that a good linear regression was established between the absorbance and the concentrations. The extracted solutions were introduced into the flame by conventional aspiration.

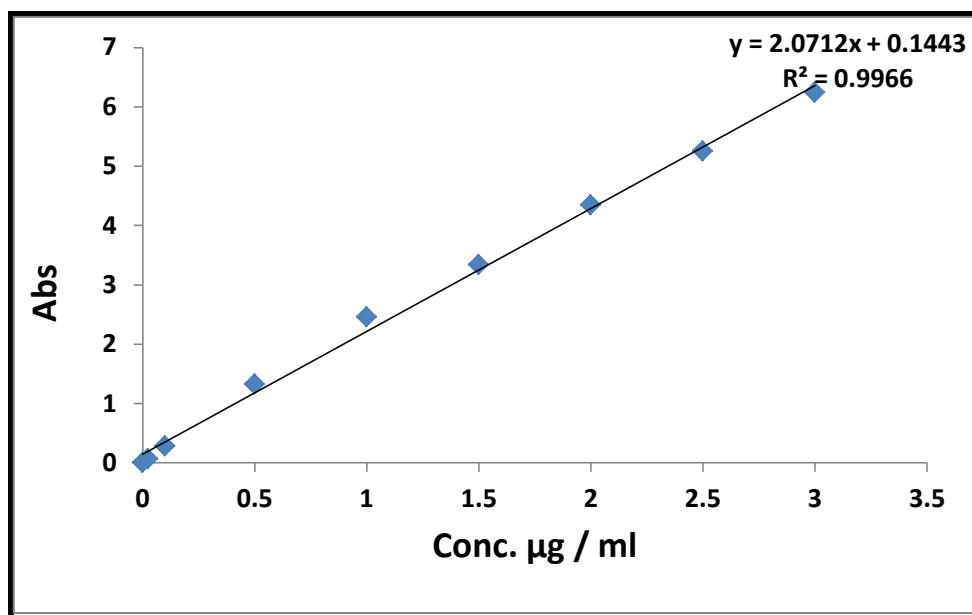


Figure (3.92): Calibration Curve for determination of cobalt ion using CPE-FAAS

3.11.6: Validation of Method and Calibration Curve Of Cobalt Ion

After completing all the optimization conditions to obtain a high sensitivity for the determination of Co^{2+} , the linear calibration curve was obtained by plotting the absorbance signal against the concentration of Co^{2+} in figure (3-92) and the data is illustrated in table (3.27). The linearity of calibration graph concentration range ($0.025 - 3\mu\text{g}.\text{ml}^{-1}$). The limit of detection (LOD) of a method is the lowest analyte concentration that produces a response detectable above the noise level of the system, typically, three times the standard deviation of the blank ($n=8$). The limit of quantitation (LOQ) is the lowest level of analyte that can be accurately and precisely measured. The limit of quantitation, defined as 10 times the standard deviation of the blank ($n=8$). The limit of detection and limit of quantitation (LOQ) were of ($0.011534, 0.038446$) $\mu\text{g}.\text{ml}^{-1}$ respectively,

depending on the standard deviation of the response and the slope of the calibration curve using the following equation: ($LOD=3\sigma_B/s$), ($LOQ=10\sigma_B/s$), where σ_B is the standard deviation of the blank calibration plot and (s) is its slope. Comparing this with the traditional method, by direct aspiration of sample, the limit of detection and the limit of quantitative were $(0.093603, 0.312010) \mu\text{g.ml}^{-1}$. The LOD and LOQ by using CPE method were 8 times smaller than the traditional. Figures (3-83) and (3-92) show these results. Furthermore, for preconcentration of 15ml of the working standard solutions based on the slope ratio of calibration curves with and without preconcentration⁽²¹⁶⁾, an enrichment factor of 54.078, was obtained for Co solution figure (3.93). To test the reproducibility of the proposed extraction method, the suggested procedure was repeated eight times under optimum conditions. The relative standard deviation (R.S.D.) was measured for $0.05\mu\text{g.ml}^{-1}$ to be 0.823%. The consumptive index is defined as the sample volume in milliliters consumed to reach a unit of enrichment factor (EF)⁽¹³²⁾: $CI = V_s(\text{ml}) / EF = 0.277$.

Table (3.27): Optimum conditions, performance characteristics for Co-L₃

No.	Parameters	Value
1	Co hallow cathode lamp element (nm)	240.72nm
2	Sample volume (ml)	15ml
3	Chelating agent (L ₃) 1*10 ⁻³ M	1ml
4	HCl	0.8M
5	Incubation time (min)	20min
6	Water bath heating temperature(°C)	60°C
7	Triton X-100 (1%)	0.9ml
8	Slope with preconcentration	2.0712
9	Slope without preconcentration	0.0383
10	Enrichment factor(Slope of CPE/Slope of Normal)	54.078
11	preconcentration factor	15
12	Consumptive index (CI)	0.277
13	Correlation coefficient (R ²)	0.9966
14	Concentration rang µg.ml ⁻¹	(0.025-3) µg.ml ⁻¹
15	Limit of detection (3σB/slope) µgml ⁻¹	0.011534
16	Limit of quantitation (10σ/slope)µg mL ⁻¹	0.038446
17	RSD% (n= 8) at 0.05µgml ⁻¹	0.823%

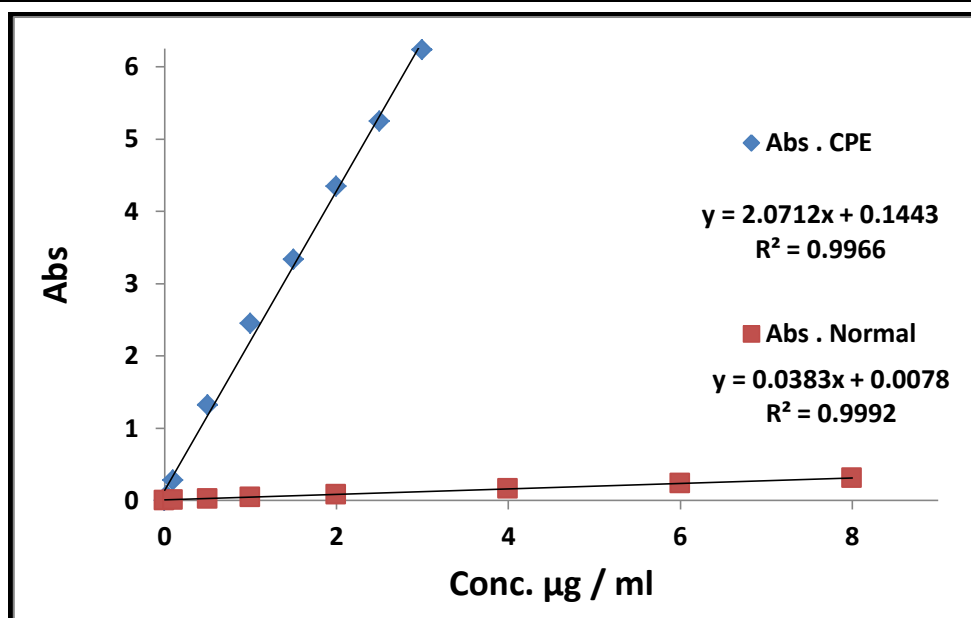


Figure (3.93): Enrichment factor between CPE method compare with normal determination

3.11.7: Determination of cobalt ion in real samples

To evaluate the effectiveness of the developed procedure for the determination of Co^{2+} ion in real samples including (milk, tobacco, tea and two different water samples including river and tap water) were applied the results as shown in figure (3.94). Good recoveries (101.108 - 104.918%) were achieved for all analyzed samples by adding Co(II) standard solution into the sample solution. FAAS was employed for the accuracy check, due to its better sensitivity. The results of this method for each type of the selected samples and the analytical recoveries are shown in the table (3.28). As it can be seen, the determination of Co^{2+} ion in real samples is noticeably accurate by the purposed method.

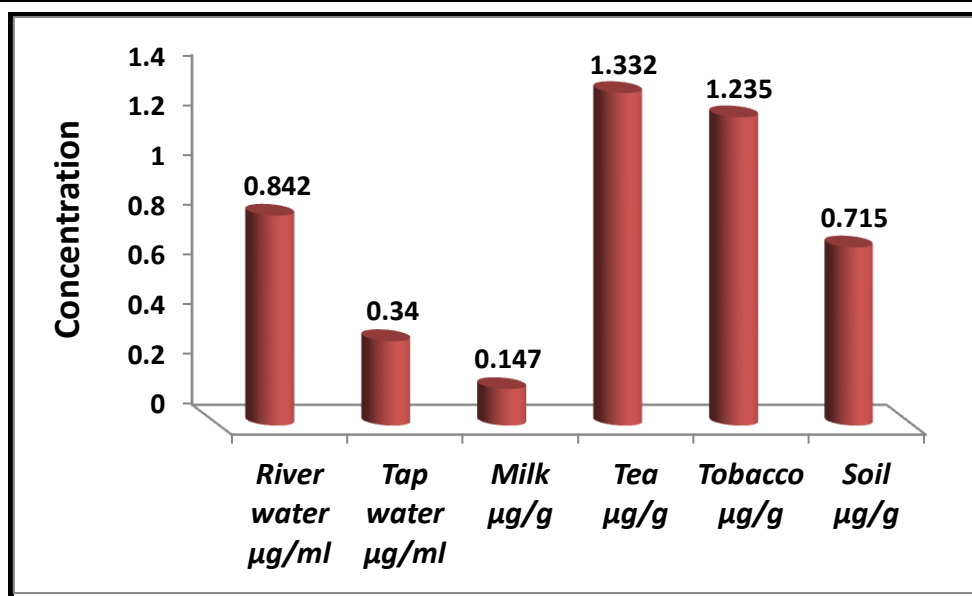


Figure (3.94): Determination of Co²⁺ ion in real samples

Table (3.28) : Recovery percentage of the cobalt ion real samples

Sample	Co Add	Co Found	% Recovery	%Error
River water ^a	0	0.842	-	-
	0.5	1.408	104.918	4.918
Tap Water ^a	0	0.340	-	-
	0.5	0.861	102.500	2.500
Milk ^b	0	0.147	-	-
	1	1.163	101.395	1.395
Tea ^b	0	1.332	-	-
	1	2.411	103.388	3.388
Tobacco ^b	0	1.235	-	-
	1	2.328	101.145	1.145
Soil ^b	0	0.715	-	-
	1	1.734	101.108	1.108

a: All values are µg.ml⁻¹, b: All values are µg.g⁻¹

The results have shown that the concentration of Co^{2+} in river and tap water was above the limit permitted concentration by WHO ($0.01\mu\text{g ml}^{-1}$) in drinking water⁽²⁴⁰⁾. There are limited reports on the concentration of cobalt in milk sample. According to a report of milk composition and synthesis resource library, the concentration of cobalt is $0.6\text{ ppm}^{(241)}$. The concentration obtained in the present study is very less compared to the reported value.

The cobalt concentration in tea was found $1.332\mu\text{g.g}^{-1}$, the determined level of Co^{2+} in the tea sample was below the limit (1.5 mg.kg^{-1})⁽²⁴²⁾. Levels of cobalt in tobacco range from <0.3 to $2.3\mu\text{g/g}$ dry weight, and approximately 0.5% of this cobalt is present in mainstream smoke cobalt. In the present study was ($1.235\mu\text{g.g}^{-1}$). The concentration of cobalt in soil may be several hundred milligrams per kilogram⁽²⁴³⁾ in the present study was below the WHO.

Conclusions

Conclusions

At the end of this thesis the points below can be concluded:

1. Compounds azo - azomethine can be prepared from azo compounds and different schiff bases.
2. Azo - azomethine compounds were able to form complexes under optimal conditions with some metal ions as an octahedral configuration and with ratio 1:1 and high values of stability constant indicated to high stability for the complexes .
3. Reagents were synthesised in this study have been proved to be an important class of reagents in analytical chemistry as organic complexing agents because of their structure features beside, their capacity to form mostly neutral and hydrophobic complexes with vast majority of metals.
4. The cloud point extraction CPE methodology is based on a group of optimal conditions that should be controlled to obtain the best extraction beside some additional factors that would change the efficiency of the extraction process.
5. The cloud point extraction CPE methodology uses a charged positive organic reagents like L_1 , L_2 and L_3 to form complexes with metals under study in order to extract and preconcentrate these complexes in the cloud point layer CP as an ion pair association complex .
6. The study shows that the acid concentration play is an important role in complexes formation and extraction within the cloud point layer CP was

[HCl] as 0.5 M for Cd (II) with L_1 , 0.3 M for Cu (II) with L_2 and 0.8M for Co^{2+} with L_3 .

7. Beside the temperature, the heating of the solution time is an important factor in determining the amount of accept heat by the solution to a process of agglomeration cloud point layer CP, and that the time to achieve the reasonable duration of a process of dehydration and layer CP conglomerate.

8. In this present study found that the newly synthesized ligands used for complexation as ion-pair association with metal of interest prior their extraction and determination by combined CPE / FAAS resulting in an increase the sensitivity, limit of detection, selectivity and enrichment factors thereby enhanced the popularity of FAAS spectrometry beside the solvent-free extraction of metals from complex matrices.

9. The results of thermodynamic study were highlighted on the behaviour of phase separation of the hydrophobic ion-pair complexes between micelles and aqueous solution. It was shown that the reaction and extraction processes are endothermic in nature which are governed by the value of ΔH . Whilst the negative ΔG value dictates that the solubilisations of the complexes studied are spontaneous phenomena and the positive value of ΔS proved that complexes molecules are organized on the mantle of hydrophobic chains a more random fashion.

10. The study of the stoichiometry for ion association complexes using the slope analysis technique showed that the ion association complex was 1:1 for all extracted complexes.

11. A success of extraction of Cd(II), Cu(II) and Co(II) by using organic reagents like L₁, L₂ and L₃ according to CPE methodology prove the possibility of the use of this technique in the estimation of the elements under study in environmental samples the results gave different high accuracy in appreciation for these elements.

12. Good recoveries (100.165-103.383%), (100.380-103.377%) and (101.108 - 104.912%) for Cd²⁺, Cu²⁺ and Co²⁺, respectively are achieved for all analyzed samples by adding standard solutions into the sample solution. FAAS was employed for the accuracy check, due to its better sensitivity.

Recommendations

Recommendations

1. The possibility of the use of the cloud point extraction technique to enrich the process of trace elements in environmental and life matrix in preparation for the appreciation according to the latest assessment techniques spectral estimation methods or other mechanism .
2. Using other nonionic surfactants such as Triton X-114, Tween 80 and OFX 0309 . These can also use industrial organic dyes as a reagents .
3. Simultaneous extraction and preconcentration for multi-elements in single sample experimental procedure of CPE is very important to shorten the analysis time and obtain much accurate results .
4. Expanded CPE methodology to metal speciation will present substantial development in contemporary analytical chemistry and open the vast ways toward easy, simple and inexpensive routine analysis instead of using expensive instrumentation in this field .
5. The continuing of preparation of new complexing agents and their exploiting in CPE will impart this method other substantial advantages especially in analytical figures of merit and simultaneous extraction of analytes .
6. Extraction and preconcentration of inorganic cation metals by CPE is also a vital theme in the future which will facilitate the determination of these species in others environmental samples with little efforts .

7. Design of CPE procedures in extraction and preconcentration of the drug will drive this method toward the determination of medicaments at trace concentration levels in wastewater in which enabling to remove these pollutants from water.

References

References

- 1- R. J. Cassella, V. A. Salima, L. S. Jesuino and R. E. Santelli, *Talanta*, 54, 61 - 67 **(2001)** .
- 2- Y. Asieh. and V. Azimi, *Eur-chem. Bull*, 2(7), 453 - 455 **(2013)** .
- 3- W. Al Zoubi, A. A. S. Al-Hamdani and Y. G. Ko, *Sep. Sci. Technol.*, 52, 1052 **(2017)** .
- 4 - W. Al Zoubi, A. A. S. Al-Hamdani and M. Kaseem, *Appl. Organomet. Chem.*, 30, 810 **(2016)** .
- 5 - W. Al Zoubi, Y. G. Ko, *App. Organomet. Chem.*, 31, 3574**(2017)** .
- 6 - N. M. Aljamal , *Biochem Anal Biochem.*, 4,2 **(2015)** .
- 7 - B. Kapil and B. Shipra, *Physics and Materials Chemistry*, 1, 1, 1- 4, **(2013)** .
- 8 - K. P. Sudhir, G. Ghosh, S. k. Rout and D. Paul, *RJC*, 6, 2, 147-152, **(2013)** .
- 9 - E. Ispir, *Dyes and Pigments*, , 82, 13 - 19 **(2009)** .
- 10 - S. Alghool, HF.A. El-Halim and A. Dahshan. *J. Mol. Struct.*, 983, 32 - 38 **(2010)** .
- 11- H. Khanmohammadi, M. Darvishpour, *Dyes and Pigments*, 81, 167-173**(2009)** .
- 12 - M. Odabas-oglu, C. Albayrak, R. Ozkanca , FZ. Aykan and P. Lonecke. *J. Mol. Struct.*, 840, 71- 89 **(2007)** .
- 13 - H. Dincapl, F. Toker, I. Durucasu , N. Avcibasi and S. Icli., *Dyes and Pigments*, 75, 11- 24 **(2007)** .

References

- 14 - H.-K. Lee, K. Doi, H. Harada, O. Tsutsumi, A. Kanazawa, T. Shiono and T. Ikeda, *The Journal of Physical Chemistry. B*, 104 , 7023 **(2000)**.
- 15 - Y. Zhang and A. Lu ZH, *Materials Chemistry and Physics*, 63,188 **(2000)** .
- 16 - A. Ghanadzadeh Gilani, M.R. Yazdanbakhsh, N. Mahmoodi, M. Moghadam and E. Moradi, *Journal of Molecular Liquids* 139 ,72 **(2008)** .
- 17 - Y.H. Ebeada, M.A. Selim, S.A. Ibrahim, *Spectrochimica Acta Part A*, 75 ,760 **(2010)** .
- 18 - V. Luksamee, L. Nararak , P. Chaveng , S. Saowanit and H. Kanidtha, *Acta Crystallogr Sect E*,1, 68 **(2012)** .
- 19 - Z. J. Mohmmmed , A. H.AL-Khafagy and A. A. M. Ali, *Int. J.Current Rese.*, 5,12, 3705-3710 **(2013)** .
- 20 - A. Nagajthi, A. Kiruthika, chitra and parameswarik., *Rese. J.Chem.Sci.*, 3,2, 35-43 **(2013)** .
- 21- Z. Huang, D. Wan, and J. Huang, *Chem Lett.*,708 - 709 **(2001)** .
- 22 - W. Kaima, N. Doslika , S. Frantza, T. Sixta, M. Wannera, F. Baumanna, *J. of Molec. Struc.*, 656, 183 - 194 **(2003)** .
- 23 - Z. Shaghaghi, *Spectrochim. Acta A*,131, 67 **(2014)**.
- 24 - M. Gulcan, S. Ozdemir, A. Dundar, E. Ispir, M. Kurtoglu and Z. Anorg. *Allg. Chem.*, 640,1754 **(2014)** .
- 25 - S. Akine, T. Nabeshima, *Dalton Trans.* 47,10395 - 10408 **(2009)** .
- 26 - H. Khanmohammadi, M. Erfantalab, A. Bayat, A. Babaei and M. Sohrabi, *Spectrochim. Acta A*, 97, 876 **(2012)** .

References

- 27 - L.D. Popov, Y.P. Tupolova, V.V. Lukov, I.N. Shcherbakov, A.S. Burlov, S.I. Levchenkov, V.A. Kogan, K.A. Lyssenko and E.V. Ivannikova, *Inorg. Chim. Acta*, 362 ,1673 - 1680 **(2009)** .
- 28 - X. Li, Y. Wu, D. Gu and F. Gan, *Dyes Pigments*, 86 ,182-189 **(2010)** .
- 29 - H. Dincálp, F. Toker, I. Durucasu, N. Avcıbası, and S. Icli, *Dyes Pigments* ,75,11-24 **(2007)**.
- 30 - T. Oktem, H. Ayhan, N. Seventekin and E. Pişkin, *Journal of the Society of Dyers and Colorists*, 115, 274- 279 **(1999)** .
- 31- A. Kakanejadifard, F. Azarbani, A. Zabardasti, S. Kakanejadifard, M. Ghasemian, F. Snaashari, S. Omid, S. Shirali and M. Rafieefar. *Dyes and Pigments*, 97, 215-221 **(2013)** .
- 32 - M. Ghasemian, A. Kakanejadifard, F. Azarbani, A. Zabardasti and S. Kakanejadifard. *Spectrochim Acta*, 124, 153-158 **(2014)** .
- 33 - H. Khanmohammadi, K. Rezaeian, *Spectrochimica Acta Part A, Molecular and Biomolecular Spectroscopy*, 97 , 652 - 658 **(2012)** .
- 34 - M. Rospenk, I. Król-Starzomska, A. Filarowski, A. Koll, *Chemical Physics*, 287,113 - 124 **(2003)** .
- 35 - F.M.W. Fabian, L. Antonov, D. Nedeltcheva, S.F. Kamounah and J.P. Taylor, *J. Phys. Chem. A*, 108, 7603 - 7612 **(2004)** .
- 36 - N. Dharmaraj, P. Viswanalhamurthi and K. Natarajan, *Transition Met. Chem.*, 26,105 - 109 **(2001)** .
- 37 - H. H. Eissa , *Journal of current research in science*, 1, 444 - 450 **(2013)** .

References

- 38 - H. Khanmohammadi and F. Khodam , *Journal of Molecular Liquids*,177, 198 - 203 **(2013)** .
- 39 - S. Bitmez , K. Sayin, B. Avar , M. Kose , A. Kayraldız and M. Kurtoglu, *Journal of Molecular Structure*,1076, 213 - 226 **(2014)** .
- 40 - M. Nic, J. Jirat and B. Kosata, eds. IUPAC Compendium of Chemical Terminology **(2006)** .
- 41 - E. Engel, H. Ulrich, R. Vasold, BurkhardKönig, M. Landthaler, R. Süttinger and W. Bäumlner, *Dermatology*, 216 ,1, 76 - 80 **(2008)** .
- 42 - K. Golka, S. Kopps, ZW. Myslak, *Toxicology Letters*,151,1, 203-10 **(2004)** .
- 43 - A. M. Al-Jasser, *Kuwait Med. J.*, 38, 171-182 **(2006)** .
- 44 - G. Ramazon, C. Tugba, I. T. Halil, *J. Chem.*, 36,1, 66 - 73 **(2012)** .
- 45 - A. Zarei, A. R . Hajipour, L. Khazdooz, B.F. Mirjalili and A. N. Chermahini, *Dyes and Pigments*, 81, 3, 240 - 244 **(2009)** .
- 46 - N. R. Jber, R. S. Abood and Y.A. Al- Dhaief , *Journal of Al-Nahrain University*, 14 ,4, 50 - 56 **(2011)** .
- 47 - N. Menek and Y. Karaman, *Dyes and Pigments*, 68,101-108 **(2006)** .
- 48 - E.H. Farouq, I.A. Essa and H.M. M. Mohanad, *Zanco*, 22,6, 78 - 87 **(2010)** .
- 49 - LI J. P , LIU P and Wang. Y. L . *Chinese Chemical Letters* ,14,7, 677 - 680 **(2003)** .
- 50 - A.A. Jarrahpour, M. Motamedifar, K. Pakshir, N. Hadi and M. Zarei , *Molecules* , 9, 815 - 824 **(2004)** .

References

- 51 - N.M. Aljamali, *J Rech*, 7, 810-838 **(2014)** .
- 52 - N. M. Aljamali, 4:2 *Biochemistry & Analytical Biochemistry*, 4,2 **(2015)** .
- 53 - N.M. Aljamali , Review in (NMR and UV-VIS) spectra As *J Rech*, 7,11**(2015)** .
- 54- W. Qin, S. Long, M. Panunzio, S. Biondi; Schiff Bases; A Short Survey on an Evergreen Chemistry Tool, *Molecules*, 18, 12264 - 12289 **(2013)** .
- 55 - S. Kumar, D.N. Dhar, P.N. Saxena; Applications of metal complexes of Schiff base-A review, *Journal of Scientific & Industrial Research*, 68 181-187 **(2009)** .
- 56 - K. Shalin, D. N. Dhar and P. N. Saxena, *J. Sci. and Ind. Res.*, 68, 181-187 **(2009)** .
- 57 - Anjani Solankee and Indrajit Thakor, *Indian J Chem*, 45b, 517 **(2006)** .
- 58 - A. Solanki and J. Patel, *Indian J Chem*, 43b, 1580 **(2004)** .
- 59 - K.R. Desai, R.B. Patel, P.S. Desai and K.H. Chikhali, *J Indian Chem Soc*,80, 138 **(2003)** .
- 60 - B. Chattopadhyay, S. Basu, P. Chakraborty, S.K. Choudhuri, A.K. Mukherjee, M.J.Mukherjee, *J. Molec. Struct.*, 932, 90 - 6 **(2009)** .
- 61- A. Xavier, N. Srividhya, *IOSR Journal of Applied Chemistry (IOSR-JAC)*,7, 06 -15 **(2014)** .
- 62 - K. Serbest, A. Özen, Y. Ünver, Er, M., I. Değirmencioğlu and K. Sancak, *J. Molec. Struct.*, 922, 1-10 **(2009)** .

References

- 63 - M.T.H. Tarafder, M.A. Ali, N. Saravana, W.Y. Weng, S. Kumar, N.U. Tsafe and K.A. Crouse, *Trans. Met. Chem*, 25,295 - 298 **(2000)** .
- 64- C. Imrie, P. Engelbrecht, C. Loubser, and C. W. McClelland, *Applied Organometallic Chemistry*,15,1,1-15 **(2001)** .
- 65 - S. Issaadi, D. Haffar, T. Douadi, S. Chafaa, D. Seraphin, M.A. Khan and G. Bouet, *Synth. React. Met-Org. & Nano-Met. Chem.*, 352 , 875 **(2005)** .
- 66 - M. Ozdemir, *Inorg. Chim. Acta*. 421,1- 9 **(2014)** .
- 67 - SK.M. Islam, A. Roy, S. Dalapati, R. Saha, P. Mondal, K. Ghosh, S. Chatterjee, K. Sarkar, N. Guchhait and P. Mitra, *J. Mol. Catal. A-Chem*. 380, 94 - 103 **(2013)** .
- 68 - S.P Netalkar, A.A. Nevrekar, and V.K. Revankar, *Catal. Lett.* 144, 1573 - 1583 **(2014)** .
- 69 - A. A. Salih and S.A.shaker, *oriental J.Chem.*, 27,3, 835 - 845 **(2011)**.
- 70 - C. Anitha, C. D. Sheela, P. Tharmaraj and V. V. Hema, *Journal of Chemistry*, Vol. 2013, ID 724163, 12 pages **(2013)** .
- 71 - Y. Arslan, et al., *TrAC Trends in Analytical Chemistry*, 30,4, 569 - 585 **(2011)** .
- 72 - Yu, C., et al., *Journal of Analytical Atomic Spectrometry*, 19,3, 410 - 413 **(2004)** .
- 73 - J. Gomez-Ariza, et al., *Analytica chimica acta*, 524,1,305 - 314 **(2004)** .
- 74 - A.N. Anthemidis and K.-I.G. Ioannou, *Talanta*, 80,2, 413 - 421 **(2009)** .

References

- 75 - S. Saçmacl, Ş. Kartal, and S. Dural, *Journal of the Brazilian Chemical Society*, 23,6,1033 - 1040 **(2012)** .
- 76 - Z. Sosa Ferrera, , C. Padrón Sanz, , C. Mahugo Santana, and J.J. Santana Rodríguez, *TrAC Trends in Analytical Chemistry*, 23,7, 469 - 479 **(2004)**.
- 77 - A.S. Yazdi , *Trends Anal.Chem.* ,30,918 - 929 **(2011)** .
- 78 - H. Watanabe and H. Tanaka, *Talanta*, 25, 585 - 595 **(1978)** .
- 79 - C.B. Ojeda and F.S. Rojas, *Microchim. Acta*, 177,1- 21 **(2012)** .
- 80 - A.A. Ghali, Z.A. Khammas, and K.H. Kadhim, *International Research Journal of Pure and Applied Chemistry*, 4,3, 362 **(2014)** .
- 81 - M. Tobiszewski, A. Mechlinska, B. Zygmunt, and J. Namiesnik, *Trends Anal. Chem.*, 28, 943 - 951**(2009)**.
- 82 - S. Armenta, S. Garrigues, and M. de la Guardia, *Trends Anal. Chem.*, 27, 497 - 511**(2008)** .
- 83 - J. Szymanowski and J. *Radioanal. Nucl. Chem.*, 246 , 635 - 642 **(2000)** .
- 84 - D. Möbius, R. Miller, and V. Fainerman, *Surfactants: chemistry, interfacial properties, applications*, Elsevier Amsterdam. p. The Netherlands, pp.1 **(2001)** .
- 85 - Z.A.A. Khammas, I.A. Ghali, PhD Thesis CH1-p 8. **(2013)**, University of Baghdad College of Science for Women.
- 86 - D. Myers,” *Surfactant science and technology* “ John Wiley & Sons, Inc. Hoboken, New Jersey.3rd Ed. p.31 **(2006)** .

References

- 87 - M.A. Rub, A.M. Asiri, M. Sheikh, K. Shafi, K. Anish, Aftab, A. P., Azum and N. Kabir ud, Din. *Journal of Molecular Liquids*, 172,0, 59 - 65 **(2012)** .
- 88 - M. d. A. Bezerra, M. A. Z. Arruda and S.L.C.Ferreira, *Applied Spectroscopy Reviews*, 40,4, 269 - 299 **(2005)** .
- 89 - F.H. Quina and W.L. Hinze, *Ind. Eng. Chem. Research.*, 38 ,4150 - 4168 **(1999)**.
- 90 - L.L. Schramm, *Surfactants: fundamentals and applications in the petroleum industry*” , 1st Ed., Cambridge University Press , UK, pp.5 **(2000)** .
- 91 - C.D. Stalikas, *Trends anal. Chem.*, 21, 343 - 355 **(2002)** .
- 92 - E. Paleologos, D. Giokas and M. Karayannis, *Trends in Analytical Chemistry*, 24,5,426 - 436 **(2005)** .
- 93 - C.B. Ojeda, F.S. Rojas and J. M. C. Pavón, *American Journal of Analytical Chemistry*,1,127-134 **(2010)** .
- 94 - C.L Liu, Y.J. Nikas, and D. Blankschtein, *Biotechnol. Bioeng.* 52 ,185-192 **(1996)** .
- 95 - K. Toerne, R. Jackson, and R. Wandruska, *J. Coll. and Interf. Sci.*, 257, 2,412 - 414 **(2003)** .
- 96 - J.A. Molina-Bolívar, C. Carnero Ruiz, *Fluid Phase Equilibria*, 327, 58 - 64 **(2012)** .
- 97 - B. Darshak, C.M. Kalpana, P. Jigisha, *J. Surfactants Deterg.*, 16, 547 - 557 **(2013)** .

References

- 98 - M. Silva, E. Cerutti and L. Martinez, *Microchimica Acta.* , 155, 349-364 **(2006)** .
- 99 - K. Pytlakowska, V. Kozik, and M. Dabioch, *Talanta*, 110 , 202 - 228 **(2013)** .
- 100 - Lenntech, Water Treatment, Published by Lenntech, Rotterdam sewage, Netherlands. Water Purification, 9, 43 - 47 **(2004)** .
- 101- P. B Tchounwou, C.G.Y., Anita K Patlolla, and J. D. Sutton, *National Center for Biotechnology Information*, 101,133 - 164 **(2012)** .
- 102 - A.R. Memon, D. Aktopraklig, Özdem and A., Vertii, *Turkish Journal of Botany*, 25, 111- 121**(2001)** .
- 103 - M. Ghaedi, R. Fathim , F. Marahhel and F. Ahamdi, *Fresen. Environ. Bull.*, 14, 1158 **(2005)** .
- 104 - A. B. El Bialy, S.S. Hamed,W.M. Moussa, and R.K. Abd El-Hameed, *Egyptian Journal of Solids*, 28,1,151-161 **(2005)** .
- 105 - T. Dauwe, E. Janssens, B. Kempenaers and M. Eens *Environmental Pollution* ,129, 125 - 129 **(2004)** .
- 106 - CFTC. Harun, and A. Ölcucu, *Int J Sci Technol*, 2, 29 - 32 **(2007)** .
- 107 - J. B. B. da . Silva, D. L. G.Borges, , M. A. M. S. da Veiga, A. J. Curtius and B. Welz, *Talanta*, 60, 977 **(2003)** .
- 108 - M. W. Ashraf and S. Akram, *Fresenius Environmental Bulletin*, 17, 7b, 877- 881 **(2008)** .

References

- 109 - S. L. C. Ferreira, J. B. Andrade, M. G. A. Korn, M. G. Pereira, V. A. Lemos, W. N. L. dos Santos, F. M. Rodrigues, A. S. Souza, H. S. Ferreira and E. G. P. da Silva, *Journal of Hazardous Materials*, 145, 3, 358-367 **(2007)** .
- 110 - Z. Krejpcio, *Journal of Environmental studies*, 10,6,399 - 404 **(2001)** .
- 111 - J. Chen, and K.C. Teo, *Analytica Chimica Acta*, 450,1, 215 - 222 **(2001)** .
- 112 - G. Vatankhah , B. Branch, Bushehr, M. Ebrahimi and M. Kahani, *Eurasian Journal of Analytical Chemistry*, 12,7, 987-1000 **(2017)** .
- 113 - Z. A. A. Khammas, I. R. Ali, S. K. Jawad, *Journal of Kufa for Chemical Science*, No. 6 ,December **(2012)** .
- 114 - M. Ghaedi, K. Niknam and M. Soylak , *pak. J.Anal . Environ. chem.*, 12 ,1,42 - 48 **(2011)** .
- 115 - A. Magda Akl, M. Magdy Bekheit and I. Helmy, *Journal of Analytical & Bioanalytical Techniques*, 7,4 **(2016)** .
- 116 - E. Kendüzler and A.R.Türker , *Chem. Acta*, 480, 259 **(2003)**.
- 117 - A.R. Ghiasvand ,R. Ghaderi and A.Kakanejadifard , *Talanta*, 62, p. 287 **(2004)** .
- 118 - E. Kenduzler and A. R. Turker, *Anal. Chim. Acta*, 480, 259 **(2003)** .
- 119 - Z. Szigeti, I. Bitter , K. Toth , C. Latkoczy , D.J. Fliegel, D. Gunther and E. Pretsch, *Anal. Chim. Acta*, 532, p. 129 **(2005)** .
- 120 - M. Tuzen , M. Soylak , D. Citak ,H.S. Ferreira, M.G.A. Korn and M.A. Bezerra , *J. Hazard. Mater.*, 162, p. 1041 **(2009)** .

References

- 121 - O.D. Sant'Ana, L.D. Jesuino, R.J. Cassella, M.B. Carvalho, and R. E. Santelli, *J. Braz. Chem. Soc.*, 14, 728 **(2003)** .
- 122 - M. Ghaedi and A. Shokrollahi, *Fresenius. Environ. Bull.*, 15, 79 **(2006)** .
- 123 - S. A. Ahmed, *J. Hazard. Mater.* 156, 521- 525 **(2008)** .
- 124 - A. S. Amin and M. A. Kassem, *TACL*, 6 ,6, 820 - 833 **(2016)** .
- 125 - Z. Yurtman-Gündüz¹, O. Aydın-Urucu¹, E. Kök -Yetimoğlu, Hayati Filik, *Macedonian Journal of Chemistry and Chemical Engineering*, 32, 1, 69 - 78 **(2013)** .
- 126 - X. Guoqiang , W. Shengping , J. Xiuming , L. Xing and H. Lijun, *J. Chem. Chem. Eng.* 30, 3 **(2011)** .
- 127 - N. Baghban, A. Mohammad H. Shabani, S. Dadfarnia and A. Ali Jafari, , *Croat. Chem. Acta* 85 ,1,85 - 90 **(2012)** .
- 128 - N. Baghban, A. M. H. Shabani, S. Dadfarnia and A. A. Jafari , *J. Braz. Chem. Soc.*, 20, 5, 832 - 838 **(2009)** .
- 129 - R. K. Sharma, M. Agrawal and F. M. Marshall, *Food and Chemical Toxicology* ,47, 583 - 591 **(2009)** .
- 130 - S. Saracoglu and M. Soylak, *Food Chem Toxicol* , 48,1328 - 33 **(2010)** .
- 131 - V. A. Lemos and G.T. David, *Microchemical Journal*, 94, 42 - 47 **(2010)** .
- 132- E. L. Silva and P.S. Roldan , *J. Hazard. Mater.*, 161, p. 142 **(2009)** .

References

- 133 - R. ELSheikh , A. Gouda, H. ELSayed and E. ALamin ,
International Journal of Pharmacy and Pharmaceutical Sciences, 7, 9
(2015) .
- 134 - S. Wang, S. Meng and Y. Guo. , *Int J Spectrosc*, 1,7 (2013) .
- 135 - D. Citak and M. Tuzen, *Food and Chemical Toxicology* , 48, 1399 -
1404 (2010) .
- 136 - X. Yang, Z. Jia, X.Yang, Li, G., and X. Liao, *Saudi Journal of
Biological Sciences*, 24,3, 589 - 594 (2017) .
- 137 - K. C. Hung, B. H. Chen, and L. E. Yu , *Separation and
Purification Technology*, 57,1, 1-10 (2007) .
- 138 - C. D. Stalikas, *Analytical Chemistry*, 21,5, 343 - 355 (2002) .
- 139 - S. Xie, M. C. Paau, C. F. Li, D. Xiao and M.M. F. Choi, *Journal
of Chromatography A*, 1217,16, 2306 - 2317 (2010) .
- 140 - Z. Wang and Z. Dai, *Enzyme and Microbial Technology*, 46,6,
407 - 418 (2010) .
- 141 - K. Sharma, P. Shivaji, R. Sandeep and A.K. Rakshit, *Colloids
and Surfaces A: Physicochemical and Engineering Aspects*, 219 ,1-3,
67-74 (2003) .
- 142 - A. Q. Shah, T.G. Kazi, J.A. Baig, H.I. Afridi, G.A. Kandhro,
M.B. Arain, and S.K. Wadhwa, *Food and Chemical toxicology*, 48,1,
65 - 69 (2010) .
- 143 - K. Materna, I. Milosz, I. Miesiac, G. Cote and J. Szymanowski,
Environmental Science & Technology, 35,11, 2341- 2346 (2001) .
- 144 - A. Shah, (2010, June 06). Water and development. Retrieved from
<http://www.globalissues.org/article/601/water-and-development>.

References

145 - S.M. Kleiner, *Journal of American Dietetic Association* 9,4 (2003) .

146 - Z. Jaric, G.Visnjic-Jeftic G, Z. Cvijanovic , L. Gacic , S. Jovanovic S, et al. *Microchemical Journal*, 98,77- 81 (2011) .

147 - Supriy Antog. Chromatomembrane Method Applied in Pharmaceuticals Analysis, Ph.D Thesis, Institute of Chemistry, Fachbereich Biologie, Chemie, und Pharmazie, Free University of Berlin, (2005) .

148 - A . M.A. Enb, A. Donia, Abd-Rabou, Abou Arab and M. H. El-Senaity, *Global Veterinaria*, 3,3, 268 - 275 (2009) .

149 - M .I Yahaya , G. C. Ezor, Y. F. Musa and S. Y. Muhamad, *African journ. Of pure and applied chem.* 4: 3(2010) .

150 - E. E. Ogabiela, G. G. Yebpella, A. F. Ade-Ajayi, U. J. Mmereole, C. Ezeayanaso, E. M. Okonkwo, D. O. Ahola, U. U. Udiba, A. Mahmood, and I. Gandu, *Journal of Pure and Applied Sciences* ,6 ,3, 325 - 331 (2010) .

151 - LI-Quang Φin, Xino – Ping Wang, Wei Li., Xing Tong and Wei-jun Tong, *Journal of Health Science* 555,2,300 - 330 (2009) .

152 - S. Binghila, D. Simona, G .Stanciu and A. Soceanu,. *Environmental Engineering and Management Journal.* 6, 805-808 (2008) .

153 - Z. Dobrzanski, R. Kalaeu, H. Gorecka, K. Cholnaka, and Barkowalk, *Polish Journal of Environmental Studies*, 14 ,5, 685 - 689 (2005) .

154 - M.R. Mosara and R.N. Roy, Guide to laboratory establishment for plant nutrient analysis. FAO fertilizer and plant nutrition bulletin, Rome, Italy. 19,1-103 (2008) .

References

- 155 - S. Verma and I. Yadav Singh, *Food Chem Toxicol* (2010) .
- 156- O. Geiss, D. Kotzias. Tobacco, cigarettes and cigarette smoke an Protection, European overview, Institute for Health and Consumer Commission (2007) .
- 157 - I.C. Nnorom, O. Osibanjo and C.G. Oji-Nnorom, *African Journal of Biotechnology*, 4,10, 1128 -1132 (2005) .
- 158 - P. Zapryanova and R. Boshinova, *Ecology and Future*, 3, 25 - 29 (2004) .
- 159 - A. Pourkhabbaz, and H. Pourkhabbaz, *Iranian Journal of Basic Medical Sciences*, 15,1, 636 - 644 (2012) .
- 160 - M.W. Ashraf, *The Scientific World Journal* 2012, Article ID 729430, 5 pages (2012) .
- 161 - RS. Pappas, GM. Polzin, CH. Watson and DL. Ashley. *Food Chem Toxicol*, 45, 202 - 209 (2007) .
- 162 - T. Karak, R. Bhagat , *Food Res Int.*, 43,9, 2234 - 2252 (2010) .
- 163 - X. Li X, Z. Z. Zhang , P. Li, Q. Zhang, W. Zhang, X. Ding , *Food Res Int*, 53,2,649 - 658 (2013) .
- 164 - B. Srividhya, R. Subramanian and V. Raj , *Int J Pharm Pharm Sci.*, 13, 257 - 258 (2011) .
- 165 - AC. Achudume and D. Owoeye , *Health* 2,09,1097 - 1100 (2010) .
- 166 - RK. Sharma, M. Agrawal and F. Marshall , *India. Ecotoxicol Environ Saf*, 66,2, 258 - 266 (2007) .

References

- 167 - L. Järup , Hazards of heavy metal contamination. *Br Med Bull*, 68,1,167 - 182 **(2003)** .
- 168 - E. Ghorbani-Kalhor, M. Behbahani, J. Abolhasani and R.H. Khanmiri , *Food Anal Methods* 8,5,1326 -133420 **(2014)** .
- 169 - M. Behbahani, J. Abolhasani, M.M. Amini, O. Sadeghi, F. Omid, A. Bagheri and M. Salarian , *Food Chem* 173,1207 - 1212 **(2015)** .
- 170 - B. Wei and L. Yang, *Microchem. J.*, 94, 99 - 107 **(2010)** .
- 171- H. Zeng, *J. Meteorol. Environ.*, 24, 52 - 55 **(2008)** .
- 172 - W.X. Liu, L.F. Shen, J.W. Liu, Y. W. Wang and S.R. Li, *Bull. Environ. Contam. Toxicol.*, 79, 209 - 213 **(2007)** .
- 173 - US - Government. Control of emissions of hazardous air pollutants from mobile sources: Final rule. *Fed. Regis.*, 66, 80 - 86 **(2001)** .
- 174 - S. Sinha, K. Pandey , A. Gupta and K. Bhatt, *Bull. Environ. Contam. Toxicol.*, 74, 210 - 218 **(2005)** .
- 175 - S. Willers, L. Gerhardsson and T. Lundh, *Respir. Med.*, 99, 1521 - 1527 **(2005)** .
- 176 - G. Nabulo, H. Oryem-Origa and M. Diamond, *Environ. Res.*, 101, 42 - 52 **(2006)** .
- 177 - J.A. Carrero, N. Goienaga, O. Barrutia, U. Artetxe, G. Arana, A. Hernández, J.M. Becerril and J.M. Madariaga, *Highw. Urban Environ.* 17, 329 - 336 **(2010)** .
- 178 - X. Bi, L. Ren, M. Gong, Y. He, L. Wang and Z. Ma, *Geoderma*, 155, 115 - 120 **(2010)** .
- 179 - X. Chen, X. Xia, Y. Zhao and P. Zhang, *J. Hazard. Mater.*, 181, 640 - 646 **(2010)** .
- 180 - J. Bai, B. Cui, Q. Wang, H. Gao and Q. Ding, *Stoch. Environ. Res. Risk Assess.*, 23, 341 - 347 **(2009)** .

References

- 181 - M. Saeedi, M. Hosseinzadeh, A. Jamshidi and S. Pajoooheshfar, *Environ. Monit. Assess.*, 151, 231 - 241 **(2009)** .
- 182 - R.T. Mehdi, E.K. Kareem and S.E. Hussien, *NJC*, 34, 361-373**(2011)** .
- 183 - E. Emin, Y. S. Eylem, K. A. Rafet and N. K. Nilgu, *Transition Met Chem*, 34,167 - 174 **(2009)** .
- 184 - Sh. M. Eassa , *J. Chem. Pharm. Res.*,8,8, 85 - 92 **(2016)** .
- 185 - K. J. AL-Adilee, K. A. Abedalrazaq and Z. M.AL-Hamdiny, *Asian J. Chem.*, 25, 18 , 10475-10481 **(2013)** .
- 186 - Z . A. A. Khammas , I. R. Ali and S. K.jawwad, *Journal of Kufa for Chemical Science* No. 6, **(2012)** .
- 187 - P. W. Atkins : Physical Chemistry. 5th Edition. Oxford University , United Kingdom. **(1994)** .
- 188 - A. M. Engida and B. S. Chandravanshi, *Chemistry International* 3,3, 212 - 218 **(2017)** .
- 189 - B. Peters (Ed.), Recommended Methods of Manure Analysis **(2003)**.
- 190 - S. M. Farid, M. A. Enani and S. A. Wajid, *-Engg. Sci.*, 15,2, 131-140 **(2004)** .
- 191 - B. Srividhya, R. Subramanian and V. Raj, *Int J Pharm Pharm Sci*, 3, 4, 257 - 258 **(2011)** .
- 192 - C.M. A. Iwegbue , G. E. Nwajei , J. E. Ogala and C.L. Overah, *Environ Geochem Health* , 32,415 - 430 **(2010)** .
- 193 - S .M. Hezaveh, H. Khanmohammadi and M. Zendehdel, *Spectrochimica Acta Part A: Molecular and Biomolecular spectroscopy*, 199,21 - 31 **(2018)**.

References

- 194 - G. Özkana, M. Kösea, H. Zenginb, V. McKeec and M. Kurtoglua, *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 5,150, 966 - 73(2015).
- 195 - A. A. Salih AL-Hamdani , A. M. Balkhi, A. Falah and S. A. Shaker, *J. Chil. Chem. Soc.*, 60,1(2015) .
- 196 - A. Ourari, K. Ouari, W. Moumeni and L. Sibous, *Transition Metal Chemistry*, 31,169 - 175 (2006) .
- 197 - K. J. AL-Adilee, S . M. Eassa and H . K. Dakhil, *Orient. J. Chem.*, Vol. 32(5), 2481-2491 (2016) .
- 198 - A. F. Husseina, R. S. Hatamb and I. M. Shaheedc, *international journal of pharmaceutical research*, 6,4 (2016) .
- 199 - W. A. Mahmoud , A. M. Ali and T. A. Kareem, *Baghdad Science Journal*, 12,1 (2015) .
- 200 - A. J. Jarad, *Journal of Al-Nahrain University*, 15,4,74 - 81(2012) .
- 201 - E. K. Kareem, *Journal of Kufa for Chemical Science*, 2,1 (2016) .
- 202 - H. H. Eissa, *J. Current Res. Sci.*, 1,2, 96 - 103 (2013) .
- 203 - Arshed Abd Ali Shihad, M.S.c thesis, p30, Eastern Mediterranean University (2013) .
- 204 - G.Yeğiner, M. Gülcan, S. Işık , G. O. Ürrit, S. Ozdemir and M. Kurtoğlu, *J Fluoresc*, 27,6, 2239 - 2251 (2017) .
- 205 - K. J. Al-adely, A. A. Mohamad Ali and R. T. Mehdi, *National Journal of Chemistry*, 38, 311(2010) .
- 206 - F. Purtaş, K. Sayin, G. Ceyhan, M. Köse and M. Kurtoglu, *Journal of Molecular Structure*, 1137, 5, 461- 475 (2017) .
- 207 - M. Sarigul, P. Deveci, M. Kose, U.Arslan, HT. Dagi, M. Kurtoglu , *J Mol Struct* ,1096,64 - 73 (2015) .
- 208 - K. J. Kadhim, M. G. Munahi, *Journal of Global Pharma Technology*, 10,9,383 - 386 (2017) .

References

- 209 - C. Demetgül , D. Deletioğlu , F. Karaca, S. Yalçinkaya , M. Timur and S. Serin, *Journal of Coordination Chemistry*, 63, 12, 2181-2191(2010) .
- 210 - Y. Hijji, B.Barare, G. Wairia, R. J. Butcher and J. Wikairad, *Acta Cryst. E*,71, 385 - 387 (2015) .
- 211- T. M. Musa¹, *International Research Journal of Pure &Applied Chemistry*,11,2,1 - 15 (2016) .
- 212 - D.A.Skoog,"Fundamental of Analytical chemistry",5th ed., New York, 87, 24 - 44, (1988) .
- 213 - R. D. Feltham and R. G. Hayter, *J.Amer.Chem.Soc.*,82,587 - 459, (1964) .
- 214 - R. Leardi , *Anal. Chim. Acta*, 652, 161- 172 (2009) .
- 215 - S. K. Jawad and E. A. Azooz , *International Journal for Sciences and Technology* , 9, 1,17 - 23 (2014) .
- 216 - I. R. Ali, M.Sc. Thesis for education college for girls, University of Kufa: p109 -115 (2005) .
- 217 - M. k. Purkait and S.S De, *Sep. Purif. Technol.* 51,137- 142(2006) .
- 218 - S. K. Jawad and L.A. Khaleel, *International Journal of Scientific Research and Modern Education (IJSRME)* , Vol. I, Issue I (2016) .
- 219 - Z . A. A. Khammas , S. K. jawwad and I. R. Ali, *Global Journal of Science Frontier Research Chemistry* ,13 ,8 (2013) .
- 220 - M. Ghaedi, A. shokrollahi, K. Niknam and M. Soylak , *pak. Separation Science and Technology*, 44, 773 - 786 (2009) .
- 221 - Q. Santos, I. Moreno, L. Santos, A. G. Santos, S. Valdinei, Souza and M. A. Bezerra , *Anais da Academia Brasileira de Ciências* , 88, 2 , 791-799 (2016) .
- 222 - World Health Organization (WHO) Guidelines for drinking water quality, (2011), fourth edition, ISBN 978 92 4 1548151.

References

- 223 - A. Ayar, D. Sert and N. Akin. The trace metal levels in milk and dairy products consumed in middle Anatolia -Turkey, *Environmental Monitoring Assessment*, 152,1-12 **(2009)** .
- 224 - Organization J fwecofawh: Safety evaluation of certain mycotoxins in food, vol. 74. *Food and Agriculture Org.* **(2001)** .
- 225 - Industrial Standards of the Ministry of Agriculture of the People's Republic of China. Residue limits for chromium, cadmium, mercury, arsenic and fluoride in tea. NY 659 - 2003, **(2003)** .
- 226 - M. Cupit, O. Larsson, C. de Meeus , GH. Eduljee and M. Hutton.. *Sci Total Environ*, 291:189e206 **(2002)** .
- 227 - F. P. Andrade, C. C. Nascentes and L. M. Costa, *J. Braz. Chem. Soc.*, 20,8 **(2009)** .
- 228 - G .N. Iwuoha, E. I. Oghu, U. I. Onwuachu, *J. Appl. Sci. Environ. Manage.* ,17 ,4, 561 - 564 **(2013)** .
- 229 - WHO Regional Office for Europe. *Air Quality Guidelines*. Copenhagen **(1987)** .
- 230 - WHO/FAO. *Codex alimentarius commission. Food additives and contaminants. Joint FAO/WHO Food Standards Programme, ALINORM* 10/12A. **(2001)** .
- 231- V.O. Doroschuk, S.O. Lelyushok, V.B. Ishchenko and S.A. Kulichenko, *Talanta*, 64, 853 - 856 **(2004)** .
- 232 - S. K. Jawad, S.M. Hameed and S. A. Hussein, *Baghdad Science Journal*, 11, 1 **(2014)** .
- 233 - I. Bushra, A. Saatea, S. Samina and K. Riaz, *British Biotechnol .J.*, 4,8, 883 - 893 **(2014)** .
- 234 - European Commission (EC). Setting maximum levels for certain contaminants in food stuffs. *Off. J. Eur. Communities*, 77, 1-75 **(2001)** .

References

- 235 - PB. Ning, CM. Gong CM, YM. Zhang , KK. Guo, J. Bai ,
Food Addit Contam B , 4, 28e33 **(2011)** .
- 236 - Industrial Standards of the Ministry of Agriculture of the
People's Republic of China. Green food-Tea. NY/T 288e. **(2012)** .
- 237 - M. Ghaedi , A. Shokrollahi , F. Ahmadi, H.R. Rajabi and M.
Soylak, *Journal of Hazardous Materials*,150 , 533 - 540 **(2008)** .
- 238 - S. K. Jawad and L. A. Khaleel, *IMPACT: Journal of Research in
Applied, Natural and Social Sciences*, 1, 2, 149 -158 **(2015)** .
- 239 - S. K. Jawad and A. S. Abed, *Chemical and Process Engineering
Research*, Vol.33**(2015)** .
- 240 - M. Tuzen¹ and M. Soy lak, *Polish J. of Environ. Stud.* 15, 6, 915 -
919 **(2006)** .
- 241 - V. Maheswara Rao and H. C. Ananda Murthy, *Int. Journal of
Engineering Research and Application* , 2248-9622, Vol. 7, 4, 76 - 84
(2017) .
- 242 - C . Valadez Vega, C. Zuniga Perez, S. Quintanar Gomez, JA.
Morales Gonzales, E. Madrigal Santillan, JR. Villagomez Ibarra .et al.
International Journal of Molecular Sciences, 12,4,2336 - 2350 **(2011)** .
- 243 - Dr James H. Kim and Dr Herman J. Gibb, Sciences International
Inc., Alexandria, Virginia, USA and Mr Paul D. Howe, Centre for
Ecology and Hydrology, Monks Wood, Huntingdon, Cambridgeshire,
United Kingdom , World Health Organization **(2006)**, 1211 Geneva 27,
Switzerland .

الخلاصة

تشكل تقنية الاستخلاص بنقطة الغيمة منذ نشأتها عام 1985، موضوعاً مهماً في الكيمياء التحليلية كتدبير واعد لفصل وإغناء الأيونات الفلزية والمركبات العضوية من مناشئ العينات المعقدة ، إذ بدأت تحتل الآن على مستوى عالٍ ، موقعاً بارزاً كبيراً ما بين طرائق الفصل الحديثة في البحث العلمي والتطبيقي ، بسبب بساطتها وسرعتها ودقتها وكلفتها الواطئة فضلاً عن انها تعد من التدابير التحليلية الصديقة للبيئة.

تشمل الدراسة الحالية ثلاث فصول رئيسية :

الفصل الاول يتضمن مقدمة عن مركبات الازو- ازوميثين ومعقداتها وعرض تقنيات الاستخلاص المختلفة والمبادئ والأفكار الأساسية والتطبيقات الممكنة لطريقة الاستخلاص بنقطة الغيمة في الكيمياء التحليلية . كما يشتمل على مراجعة موجزة حول التطورات الحديثة لهذه الطريقة وتطبيقاتها في تقدير العناصر الكاديوم والنحاس والكوبلت بواسطة CPE .

الفصل الثاني يتضمن استعراض شامل للأجهزة التحليلية والعامّة والمواد الكيميائية المستخدمة في هذه الدراسة الحالية. كما تم وصف المسارات التحضيرية لمركبات الازو-ازوميثين (L_1-L_4) وخصائصه والاختبارات الأولية لتكوين المعقد بين ايونات الفلزات والكواشف ، تحديد الطول الموجي الاعظم λ_{max} . فضلاً عن تحديد الظروف التجريبية المثلى (تأثير حجم الكاشف ، تأثير زمن التفاعل ، تأثير درجة الحرارة ، تأثير pH) وتحديد درجة التفكك وثابت الاستقرار والاتحاد العنصري للمعقد بطريقة التغير المستمر (طريقة جوب). بالإضافة الى معقدات تم تحضيرها L_2 مع املاح $CoCl_2.6H_2O$ و $CuCl_2.2H_2O$ و L_4 مع املاح $CdCl_2.H_2O$ و $CrCl_3.6H_2O$. فضلاً عن تفاصيل كاملة للتدابير التحليلية لطريقة الاستخلاص بنقطة الغيمة المخططة لتقدير العناصر المختارة بما في ذلك (تأثير تركيز الكاشف ، تركيز حامض HCl ، زمن الحضان (التسخين) ، درجة حرارة التوازن والدراسة الثرمودينمية وكمية التريتون وكذلك دراسة الاتحاد العنصري لمعقد الترابط الايوني المستخلص باستخدام تقنية تحليل الميل) ، تم تطبيق طريقة CPE الحالية لتحديد تركيز ايونات الكاديوم والنحاس والكوبلت في عينات حقيقية بواسطة جهاز الامتصاص الذري اللهب.

الفصل الثالث تضمن تشخيص مركبات الازو- ازوميثين ومعدقاتها بتقنيات تحليلية متنوعة مثل طيف الاشعة المرئية وفوق البنفسجية ، طيف الاشعة تحت الحمراء، الرنين النووي المغناطيسي، طيف الكتلة والتوصيلية المولارية للمعدقات وكانت النتائج مطابقة لما هو متوقع عمليا وكان الشكل الفراغي هو ثماني السطوح والاتحاد العنصري للمعدد بنسبة 1:1.

الفصل الثالث ايضا يُلقي الضوء على الجزء المُهم لهذه الدراسة والذي يتناول النتائج والمناقشة في التطبيقات التحليلية في الفصل والإغناء للفلزات باستخدام استخلاص نقطة الغيمة كتقنية استخلاص تُستخدم المواد السطحية بديلاً عن المذيبات العضوية لفصل الطور.

في الجزء الاول من استخلاص نقطة الغيمة استخدم الكاشف التحليلي (L_1) بعد التحضير والتشخيص بتقنيات تحليلية متنوعة كخطوة اولية تجاه استخدامه ككاشف عضوي لعمليات الاستخلاص لتكوين معدد ترابط ايوني (كاره للماء) مع الكادميوم على هيئة ايون سالب $CdCl_4^{2-}$ في الوسط الحامضي ملائم للتفاعل مع المادة السطحية في المحلول لفصل واغناء الكادميوم بتقنية استخلاص نقطة الغيمة ليتم استخلاصه في الحجم الصغير من الطور الغني بالسطح على هيئة $CdCl_4^{2-}$ ، ووفقاً لذلك ، تم تصميم ودراسة تدبيرين تحليليين للاستخلاص بنقطة الغيمة لتقدير الكادميوم على هيئة $CdCl_4^{2-}$ طيفياً جنباً لجنب لفهم آلية التمدوب وفصل الأطوار من خلال الدراسات الترمودينمية الشاملة لهذه الأنواع في منظومة الاستخلاص بنقطة الغيمة. أُجريت تحريات تجريبية مُختلفة للوصول الى الظروف المثلى لتحليل الكادميوم من خلال دراسة تأثير العوامل المؤثرة على كفاءة الاستخلاص لهذا العنصر باستعمال العوامل تتابعياً (OFAT)، كتأثير تركيز الكاشف وتركيز حامض الهيدروكلوريك وزمن التسخين ودرجة حرارة نقطة الغيمة وكمية المادة السطحية ، تناولت الدراسة بشكلٍ واسع ايضاً على استخراج المعلومات الترمودينمية للمساعدة في تفسير عمليات الاستخلاص والتمدوب لهذه المعدقات في المذيبات (طور المادة السطحية) وبتطبيق الظروف المثلى أمكن الحصول على عامل إغناء بمقدار 51.259 للكادميوم مع L_1 ، أما المدى الخطي للكادميوم كان (0.01-1.3) ميكروغرام مل⁻¹ وحد كشف بمقدار (0.012464) ميكروغرام مل⁻¹ ، كما أمكن تحقيق دقة محسوبة على أساس الانحراف القياسي المئوي لثمان قياسات مُتكررة عند تركيز 0.05 ميكروغرام مل⁻¹ مقدارها (1.308%) ، طبقت الطريقة المقترحة لتقدير تركيز الكادميوم في نماذج حقيقية مختلفة مثل ماء نهر وماء حنفية والحليب والشاي والتبغ والتربة . تم تحديد الدقة من خلال نسب الاستخلاص للنماذج الحقيقية حيث وجدت نسب جيدة (100.165-103.383 %) للكادميوم مع L_1 .

في الجزء الثاني من استخلاص نقطة الغيمة استخدم الكاشف التحليلي (L_2) بعد التحضير والتشخيص كخطوة أولية تجاه استخدامه ككاشف عضوي لتكوين معقد ترابط أيوني (كاره للماء) مع النحاس في الوسط الحامضي ملائم للتفاعل مع مادة الشد السطحي تحت الظروف المثالية. بعدها جرت اذابة الطور الغني للمادة السطحية الذي يحتوي على معقد النحاس بمذيب وتم قياس محتوى النحاس بواسطة جهاز الامتصاص الذري اللهبى . تأثيرات متنوعة تؤثر على كفاءة الاستخلاص بنقطة الغيمة لهذا العنصر باستعمال تدبير الأمثلية الكلاسيكي (OFAT) تتابعياً، وأُردفت بمتغيرات أخرى مهمة كالتى ذكرت في طريقة الكاديوم أعلاه. اشتمل الهدف كذلك على الدراسة الترمودينمية بغية التوسع لفهم آلية التمدوب للنحاس في المذيبات. كان عامل الإغناء تحت الظروف المثلى هو 52.574 كان ينجز للنحاس مع الكاشف L_2 ، اما المدى الخطي لمنحني المعايرة للنحاس فقد كان (3-0.01) ميكروغرام مل⁻¹ الذي ادى على الحصول على حد كشف 0.017238 ميكروغرام مل⁻¹، كما أمكن تحقيق دقة محسوبة على أساس الانحراف القياسي المئوي لثمان قياسات مُكررة عند تركيز 0.05 ميكروغرام مل⁻¹ مقدارها (1.063%) ، طبقت الطريقة المقترحة لتقدير تركيز النحاس في نماذج حقيقية مختلفة . تم تحديد الدقة من خلال نسب الاستخلاص للنماذج الحقيقية حيث وجدت نسب جيدة (100.380-103.377 %) للنحاس مع L_2 .

في الجزء الثالث من استخلاص نقطة الغيمة استخدم الكاشف التحليلي (L_3) بعد التحضير والتشخيص كخطوة أولية تجاه استخدامه ككاشف عضوي لتكوين معقد ترابط أيوني مع الكوبلت في وسط حامضي ليتفاعل مع المادة السطحية في المحلول. الكاشف (L_3) يستعمل لتكوين معقد الترابط الايوني واستخلاصه بالمادة السطحية (Triton X-100) تحت الظروف المثلى لتحديد الكوبلت على هيئة $CoCl_4^{2-}$ باستعمال استخلاص نقطة الغيمة . بعدها تجرى اذابة الطور الغني للمادة السطحية الذي يحتوي على معقد بمذيب 1 مولاري من حامض النتريك في الميثانول وتم تقدير الكوبلت بواسطة جهاز الامتصاص الذري اللهبى. تم دراسة كافة الظروف المثلى لعمليات تفاعل التعقيد والاستخلاص ، لقد وجد ان عامل الاغناء والتركيز المسبق 15 و 54.078 على التوالي ، اما المدى الخطي لمنحني المعايرة للنحاس فقد كان (3-0.025) ميكروغرام مل⁻¹ الذي ادى على الحصول على حد كشف 0.011534 ميكروغرام مل⁻¹، كما أمكن تحقيق دقة محسوبة على أساس الانحراف القياسي المئوي لثمان قياسات مُكررة عند تركيز 0.05 ميكروغرام مل⁻¹ مقدارها (0.832%) ، طبقت الطريقة المقترحة لتقدير تركيز النحاس في نماذج حقيقية مختلفة.

تم تحديد الدقة من خلال نسب الاستخلاص للنماذج الحقيقية حيث وجدت نسب جيدة (101.108-104.918 %) للكوبلت مع L_3 .

أظهرت دراسة الاتحاد العنصري لكل معقدات الترابط الايوني باستخدام طريقة تحليل الميل كانت بنسبة 1:1 لكل المعقدات المستخلصة.



وزارة التعليم العالي والبحث العلمي

جامعة ذي قار

كلية العلوم

قسم الكيمياء

تقدير بعض العناصر الثقيلة في نماذج حقيقية باستعمال مركبات أزو-أزوميثين جديدة بواسطة أستخلاص نقطة الغيمة

اطروحة

مقدمه الى مجلس كلية العلوم/ جامعة ذي قار

كجزء من متطلبات نيل درجه الدكتوراه فلسفة في علوم الكيمياء/ الكيمياء التحليلية

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