ADDIS ABABA UNIVERSITY

SCIENCE FACULTY

SCHOOL OF GRADUATE STUDIES

CHEMISTRY DEPARTMENT

Graduate Project (Chem.774)

DERIVATIZATION OF NINHYDRIN WITH ETHYLENEDIAMINE-COMPLEXATION WITH Ni(II)-SYNTHESIS AND CHARACTERIZATION

BY

MINBALE ADMAS TESHAGER

July, 2008

Addis Ababa

Derivatization of Ninhydrin With Ethylenediamine-Complexation With Ni(II)-Synthesis and Characterization

Graduate Project (Chem.774)

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By

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APPROVED BY THE EXAMINING BOARD	SIGNATURE
PROF. NEGUSIE RETTA (ADVISOR)	--------------
PROF. V. J. T. RAJU (EXAMINER)	---------------

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LIST OF ABBREVIATIONS AND SYMBOLS

Salen→ N,N′-ethylene bis (salicyliden imine)
En→Ethylenediamine
Nin→Ninhydrin
Rp→Ruhemann′s purple
IR→Infrared
UV-Vis→Ultraviolet-Visible
nm→NanoMeter (10^{-9}m)
cm^{-1}→centimeter inverse
λ→Wave Length
M.Pt→Melting point
M-L→Metal-Ligand
KBr→Potassium bromide
DMF→Dimethylformamide
DMSO→Dimethylsulfoxide
ΛM→Molar conductance
Xg→Gram Susceptibility
Xm→Molar Susceptibility
µ→Magnetic Moment
µeff→Effective magnetic moment
v→Stretching Vibration
δ→Bending Vibration
^{0}C→Degree centigrade
S→Siemens
%→Percentag

ABSTRACT

A tetradentate Schiff base ligand was formed by the 1:2 molar condensation of Ethylenediamine and Ninhydrin and Characterized by elemental analysis, IR and UV-Vis. Spectra. Its Ni(II) complex was synthesized and characterized by analytical studies, molar conductivity, magnetic susceptibility, IR and UV-Vis. Spectra. The metal to ligand ratio is 1:1 and the metal complex is a non-electrolyte. Based on the spectral data and magnetic susceptibility measurement, dibasic ONNO donor behavior of the ligand and octahedral geometry for the brick-red colored complex have been concluded.

Key Words : **Ninhydrin, Ethylenediamine, Tetradentate, Schiff base,**
Chelating Ligand, Ni(II) Complex,
Synthesis, Characterization.

1. INTRODUCTION

The present investigation deals with derivatization of Ninhydrin with Ethylenediamine and complexation of the derivative with one selected transition metal ion (Ni^{2+}). The derivative is expected to be a Schiff base and a multidentate or chelating ligand. This introductory presentation includes a brief discussion on the related subjects.

Metal complexes of biologically active and structurally significant ligands provide valuable bonding and applicational understanding[14]. In this study precursors for ligand synthesis were carefully identified to generate such an understanding. Biologically active molecules and their active derivatives invariably function through multiple binding centres. When the available electron donating or binding centres are incompletely utilized in metal ion coordination, the resultant metal complex(es) can cause modified electronic distributions in the ligand and also the biological properties. This project is attempted under this broad perspective. However, the time frame has permitted an investigation only on the synthesis and characterization of the compounds.

1.1. Multidentate NO-Donor Ligands And The Chelate Effect

Ligands that can be attached to a central metal through more than one atom are said to be multidentate ligands (chelating ligands) and their complexes are called Chelates, the number of actual coordinating sites being indicated by the terms bidentate, tridentate, tetradentate, and so on[1] .

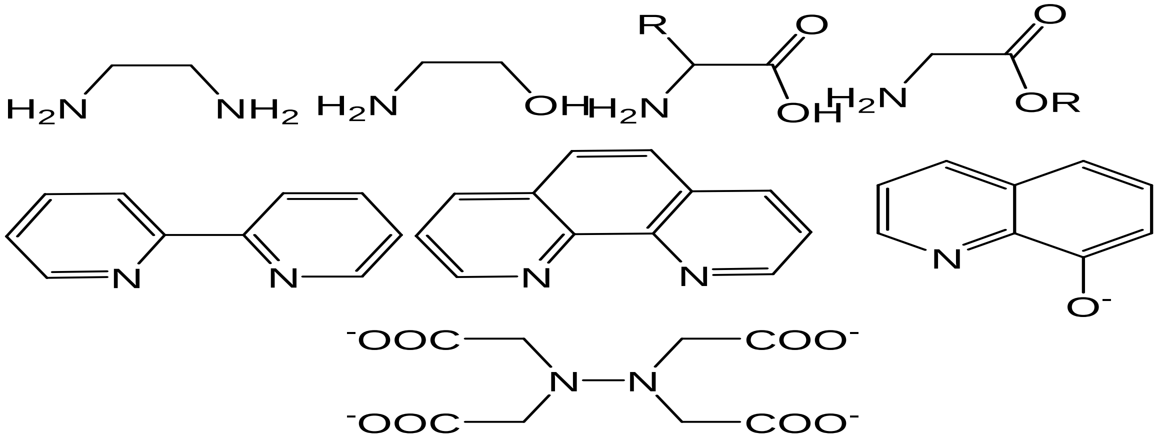


Figure -1: Some typical NO-donor chelating Ligands

Complexes of chelating ligands are in general more stable thermodynamically than those with an equivalent number of monodentate ligands. This special effect is known as chelate effect , an effect which can be explained as arising from a large positive entropy change when two or more monodentate ligands are replaced by one[1,2].

$$ML_n + (LL)_{n/2} \rightleftharpoons M(LL)_{n/2} + nL$$

Where, L=Monodentate Ligand & LL=Bidentate Ligand
The Equilibrium lies to the right

Reactions logk

$$Ni(NH3)_2 2+ \ +en \rightleftharpoons Ni(en)^{2+} + 2NH_3 \ \text{-------}2.41$$

$$Ni(NH3)_4 2+ \ +2en \rightleftharpoons Ni(en)_2^{2+} + 4NH_3 \ \text{----}3.31$$

$$Ni(NH3)_6 2+ \ 3en \rightleftharpoons Ni(en)_3^{2+} \ +6NH_3 \ \text{---}3.56$$

Sheme-1: The chelate effect and Thermodynamic Stability

The above scheme can explain the chelate effect and thermodynamic stability of the $Ni(en)_3^{2+}$ complex, increasing the formation constant(logk) value indicates increasing stability ,due to the number of species increased from 4 to 7 on the product side , which results an increase in entropy, an increase in entropy makes the formation of the chelated complex more favorable. The size of the chelating ligands are also important. Generally 5-membered chelates are more stable than 6-membered ones which are in turn more stable than 7- membered, so that ligands like en, oxalate and glycinate form stable 5-member chelates[1].

With the same size of chelate rings and with the same metal ion the logk values will usually in the order of $(O\text{-}O)^{2-} < (O\text{-}N)^{-} < (N\text{-}N)$. i.e diamine complexes will be more stable than amino acid complexes which in turn be more stable than dicarboxylate complexes.

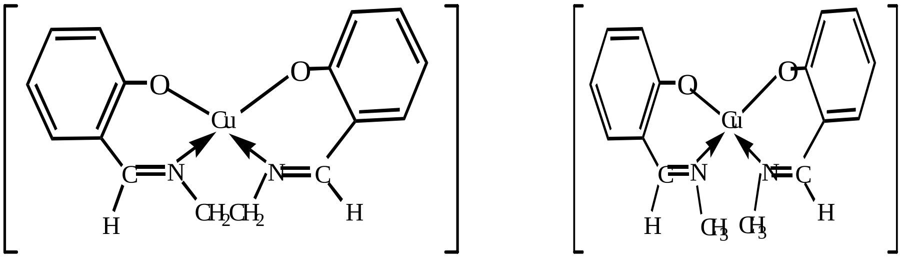
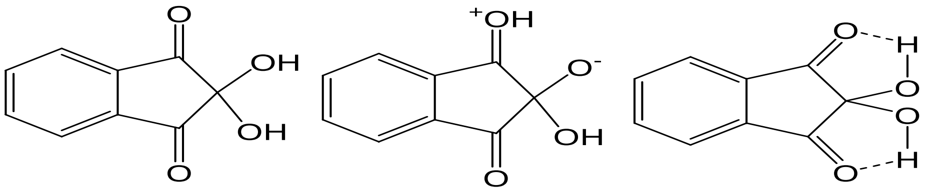


Figure-2: N,N′-ethylene bis(salicyliden imine) and N,N′-dimethyl bis(salicyliden imine) Cu(II) Complex.

In general, these species are said to be "entropy stabilized" since the replacement of two unidentate ligands by a single ligand increases the total number of ions or molecules in the system, and thus gives a positive increase in entropy[1-4].

1.2. The Chemistry of Ninhydrin

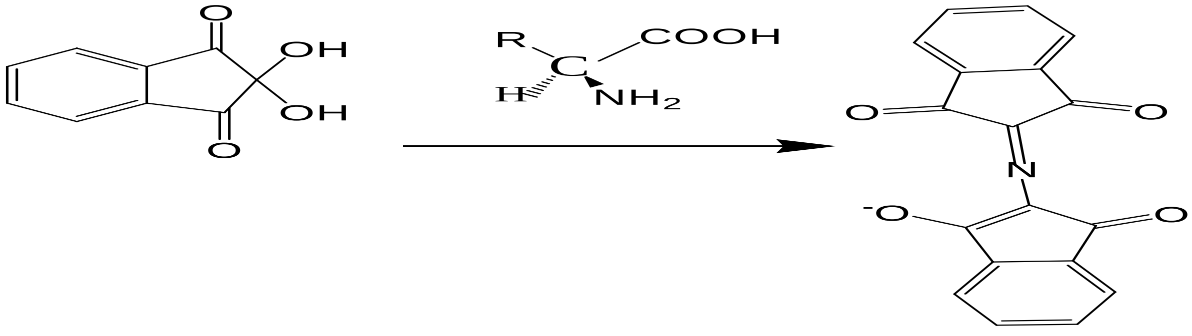
Ninhydrin was first made in 1910 by an English chemist Siegfried Ruhemann, who also investigated its reaction with amines and amino acids to form a colored compound. The product of this reaction is a compound known as Ruhemann's purple (Rp), which has an absorption maxima at 570 nm. On the basis of his experimental work, Ruhemann proposed 2,2-dihydroxy-1,3-indandione (I) as the structure of Ninhydrin. Since Ninhydrin was colorless, the keto groups in the 1- and 3- positions must be separated. To explain the the ready solubility of Ninhydrin in water and its thermal stability, Schonberg and Moubasher suggested an inner-salt or zwitter-ion formula(II). A structure which is stabilized by five-membered H-bonding is also possible(III)[5].



I II III

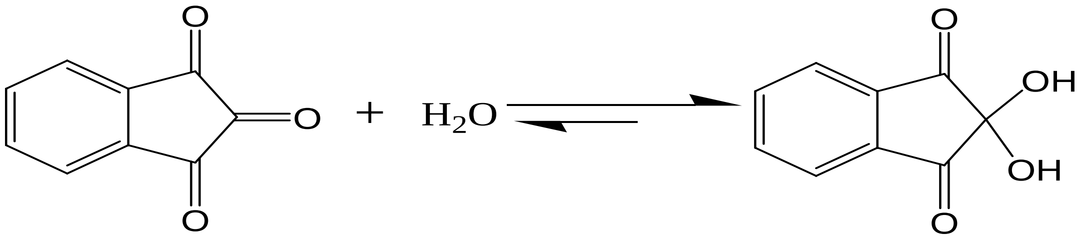
2,2-dihydroxy-1,3-indandione An inner-salt(zwitter-ion) H-bond stabilized

Figure-3: Different structures of Ninhydrin



Scheme-2 : Ruhemann's purple (Rp) formation reaction

Ninhydrin is a stable hydrate form of its parent carbonyl compound (trione). The strong electron withdrawing groups on α-C(alpha carbon) destabilizes an adjacent carbonyl group because of repulsion of adjacent positive charges. Hydrate formation overcomes the forces of repulsion. Therefore, the hydrate of the middle carbonyl group of Ninhydrin removes both pairs of repulsions and only in highly anhydrous condition exists in its trione form [5-8] .



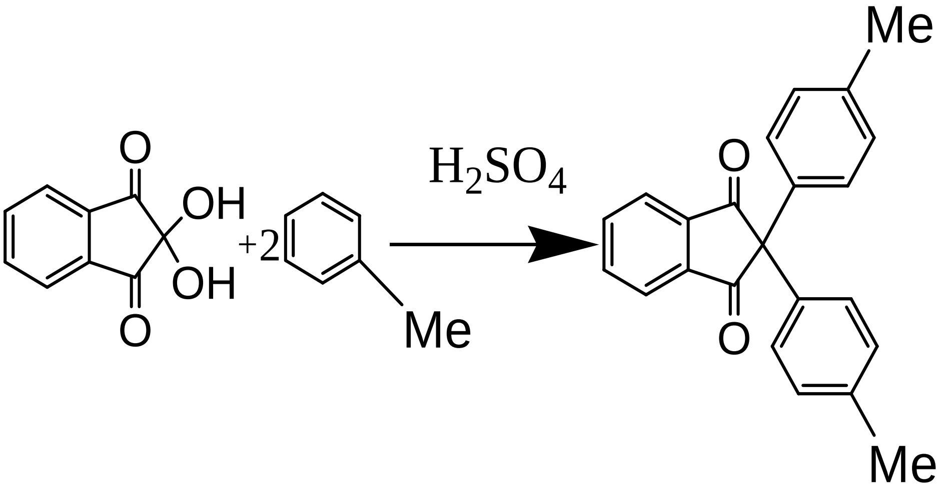
Scheme-3: Hydrate formation reaction of Ninhydrin

1.2.1. Reactions of Ninhydrin

Ninhydrin reacts with different compounds to give different products and the reactions are condition and solvent dependent, some of them are shown below[9-12].

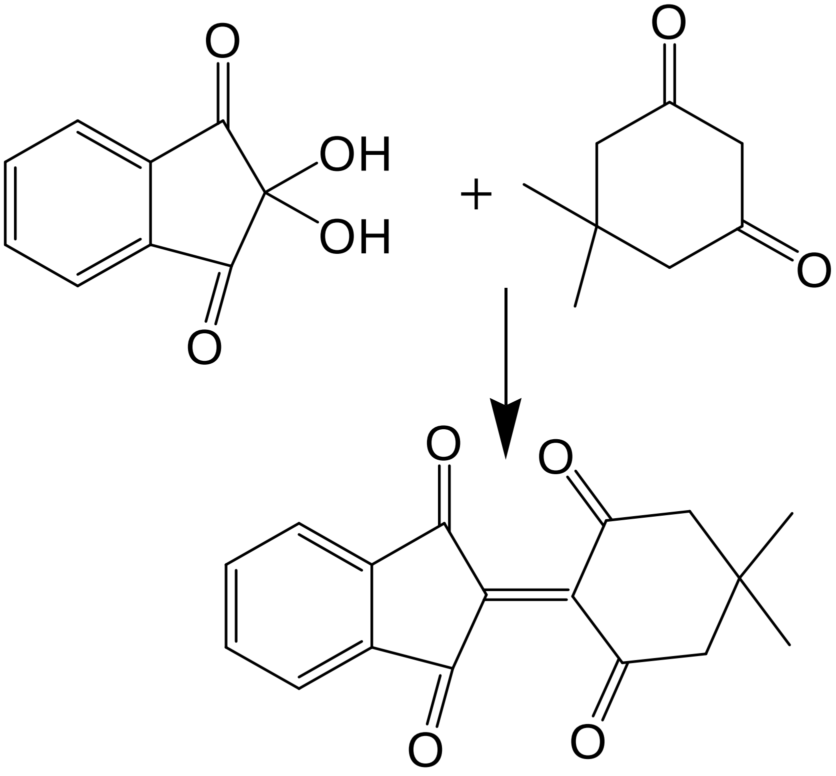
I.Reaction With Aromatic Compounds

Ninhydrin , the 2,2-diol or hydrate of indanetrione, reacts with aromatic compounds to form 2,2-diarylindane-1,3-diones: the reaction is catalysed by sulfuric acid. For example by taking toluene as aromatic reactant, the product has both methyl groups at *para-positions*[7].



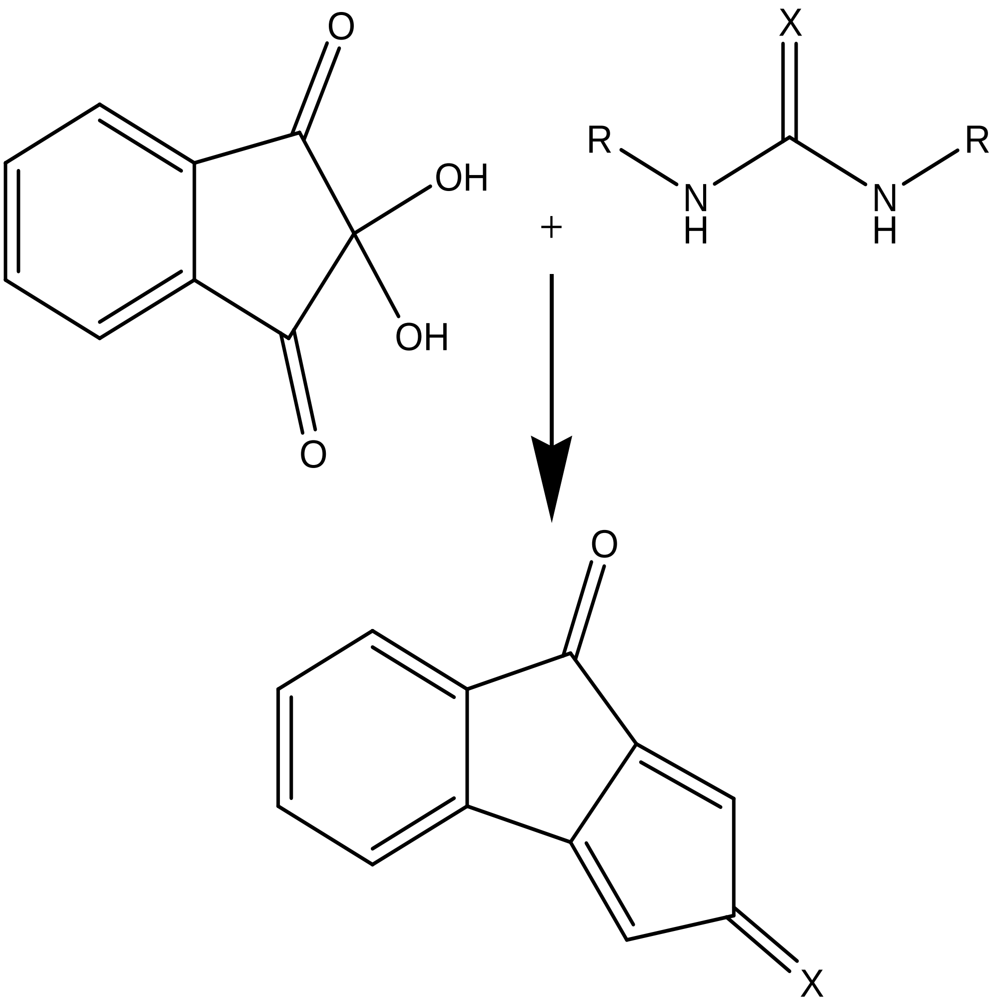
Scheme-4: Reaction of Ninhydrin with Toluene

II. Reaction With Dimedone[24]



Scheme-5: Reaction of Ninhydrin With Dimedone

III. Reaction With Thio(Urea) [24]



Where X=O, R=H

X=S, R=H

X=O, R=Me

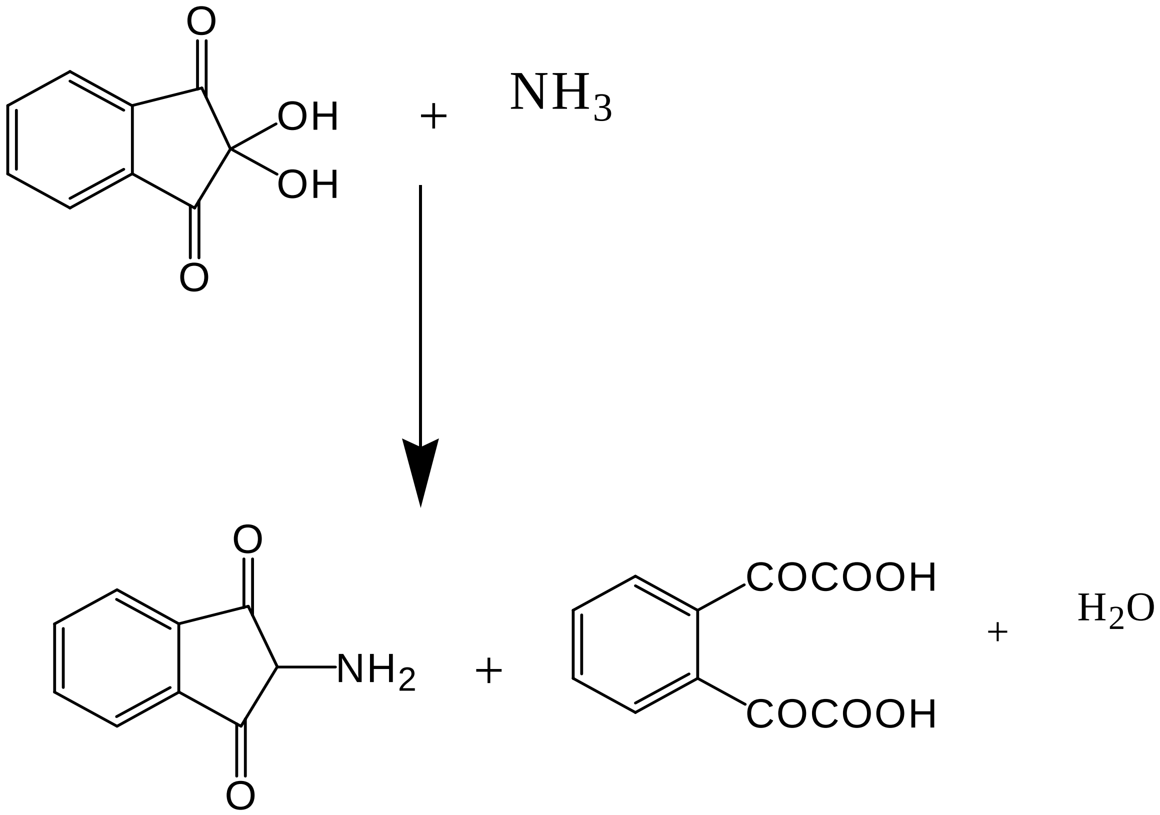
X=S, R=Me

X=S, R=Ph

Scheme-6: Reaction of Ninhydrin With Thio(Urea)

V. Reaction With Ammonia

Ammonium salts, dilute ammonia solutions, and certain amines also give a blue color under certain conditions, apparently because of an intermolecular oxidation and reduction of the ninhydrin in the presence of ammonia[7,39].

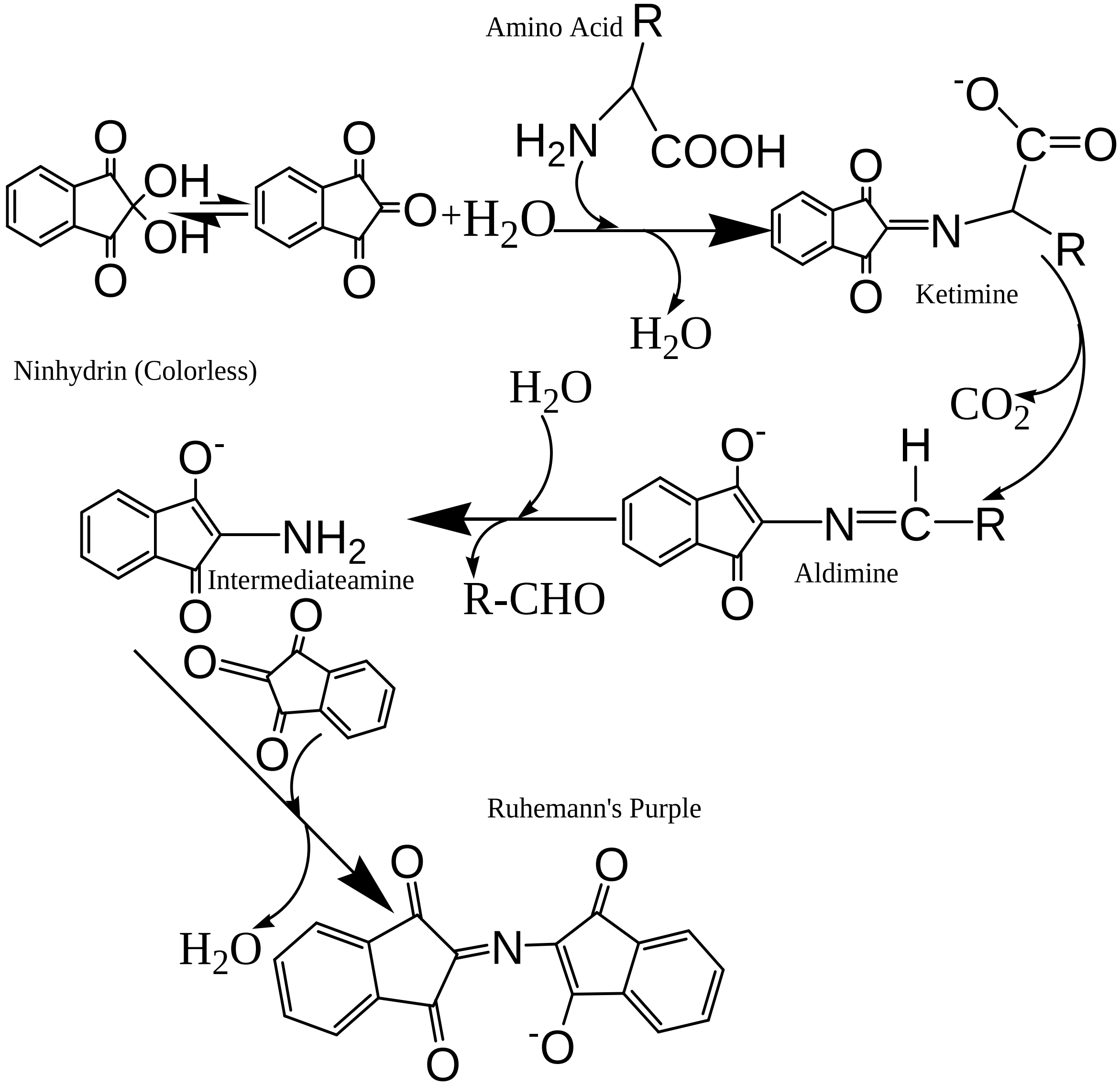


Scheme-7: Intramolecular redox reaction of ninhydrin in the presence of ammonia

VI. Reaction With Amino Acids

Ninhydrin reaction is used to detect the presence of amines, amino acids and proteins containing free amino groups. When heated with ninhydrin, these molecules give characteristic deep blue color (or occasionally pale yellow**).** The chemistry of the reaction of ninhydrin with amino acids has been studied by several workers.

The mechanism however was not well understood and this gives rise for a series of theories as reviewed by McCaldine. A simplified form of the mechanism proposed by Filippovich and McCaldin is shown in (scheme-8). It has a condensation step that leads to a Schiff base formation followed by decarboxylation, hydrolysis and finally further condensation with another ninhydrin molecule to give the final product, Ruhemann's purple *[5-8,12,14,37-40]*.



Scheme-8: The reaction of Ninhydrin with α -Amino Acids

Among the reaction products of Ninhydrin with amino acids the ketimine is a potential Schiff base ligand that can act as a tridentate ligand forming two stable five member rings on complexation. As Studies indicate the coordination of Schiff bases derived from Ninhydrin with different amino acids (glycin, L-valine, L-Alanine, L-Histidine) with some transitional metal ions (Ni^{2+}, Co^{2+}, Zn^{2+}) showed that if a metal ion is present before the reaction begins , the reaction does not always proceed to the final product (Ruhemann's purple), but stops at the first step (at ketimine stage) and the metal ions form highly stable complexes with the Schiff bases. Most of these metal complexes have octahedral geometry in which the ligands behave as monobasic tridentate (ONO) donors[7,14,39] .

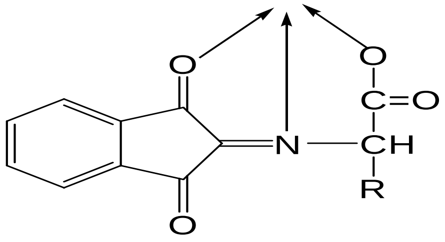
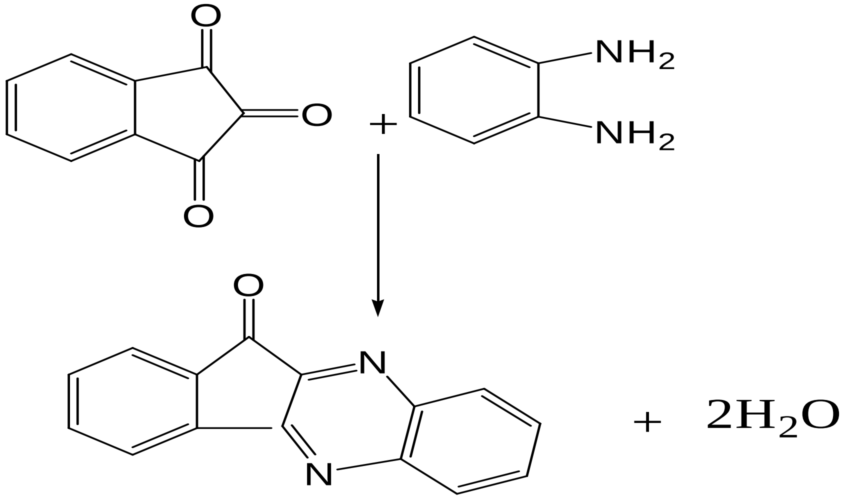


Figure-4: The Binding Sites of ketimines of Amino Acids

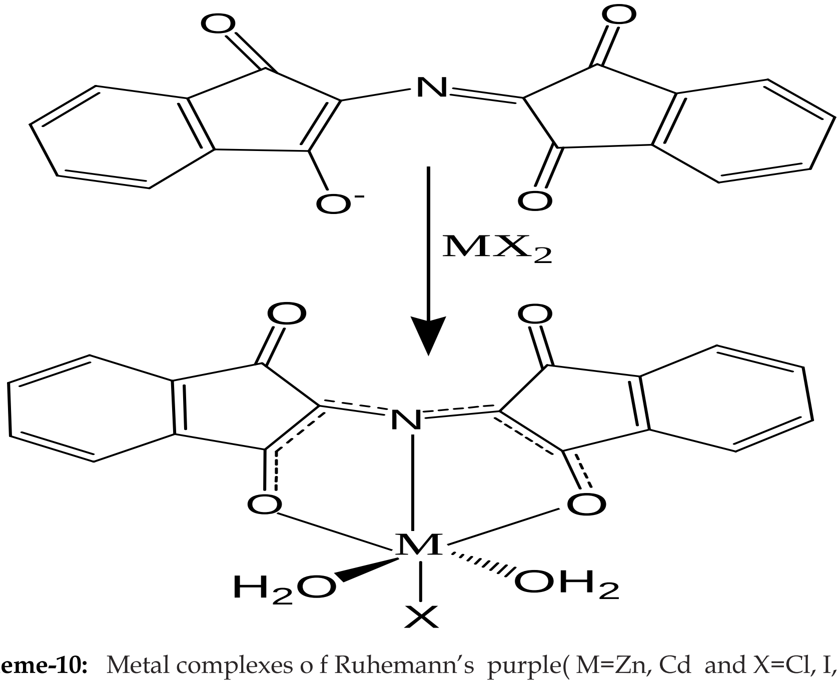
VII. Reaction With Diamines[24,40]



Scheme-9: The reaction of Ninhydrin with O- phenylenediamines

1.2.2. Metal Complexes Of Ruhemann's Purple

The reaction of metal salts with Ruhemann's purple to provide products that have strong luminescent properties has been essential in the detection of weak ninhydrin prints. The resulting complex between Ruhemann's purple and metal salts has been a subject of growing interest. It has been shown that both Zn(II) and Cd(II) salts complex with Ruhemann's purple to provide luminescent products with slightly different properties (Scheme 10). The complexes formed are very sensitive to the development conditions and can form species with either 1 : 1 or 1 : 2 metal : ligand ratio. In polar solvents it has been shown that a red 1 : 2 metal : ligand complex is formed while in less polar solvents the formation of an orange 1 : 1 metal : ligand complex is favored. The complexes each show different emission spectrum that may be beneficial depending on the background present. On paper surfaces it has been shown that products are mainly determined by the concentration of Ruhemann's purple. Strong fingerprints induced the formation of a 1 : 2 complex while weak fingerprints favored the formation of a 1 : 1 complex[12] .



Scheme-10: Metal complexes o f Ruhemann's purple(M=Zn, Cd and X=Cl, I, NO_3)

1.2.3. Applications Of Ninhydrin

Ninhydrin has found wide utility as a reagent for the spectrophotometric determination of amino acids as well as primary and secondary aliphatic amines. Aromatic amines are known to give colored condensation products with Ninhydrin. Most of these compounds have color intensity so low that the reaction is usable only as a spot test if the aromatic amine is present in high concentration. Of the compounds investigated only P-phenylenediamines and p-aminophenols give colors sufficiently intense for the spectrophotometric determination of trace amounts. The reaction of Ninhydrin is applicable for the detection, isolation and analysis of numerous compounds across a broad spectrum of disciplines such as agricultural, biochemical, medicinal, nutritional, plant science and in forensic science to study fingerprints. In line with the study of fingerprints different ninhydrin analogues were synthesized by different chemists with better sensitivity some of them are:

5-methoxyninhydrin(1), benzoninhydrin(2), thienoninhydrin(3),naphthoninhydrin(4) and in addition bis-ninhydrin(5) and Aryl and heteroaryl ninhydrin(6) analogues were also synthesized [6,7,12,14-17, 37-40] .

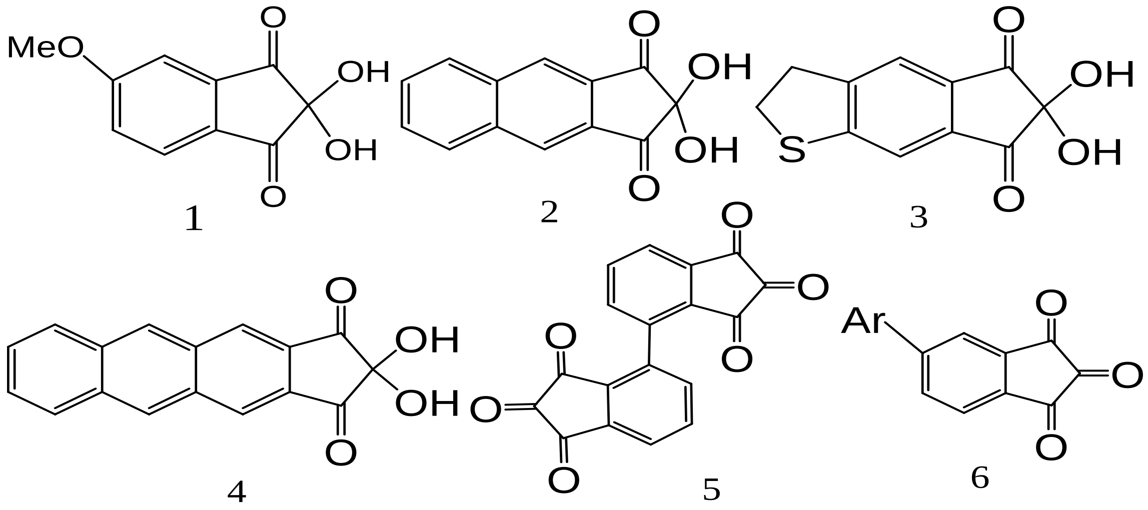


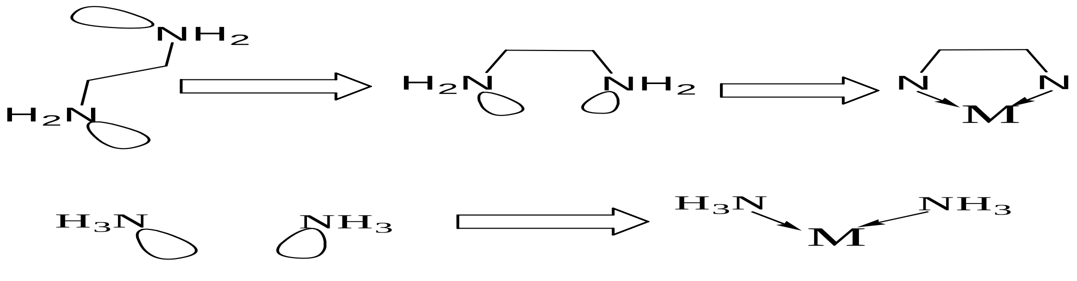
Figure-5: Ninhydrin analogues which are used in fingerprints

1.3. Ethylenediamine and Its Coordination Mode

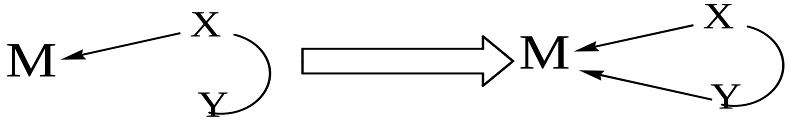
Ethylenediamine is a colorless liquid at room temperature having the characteristic smell of an amine.It is completely miscible in water and in the majority of organic solvents. In aqueous solution two equilibria are possible as shown below[38].

$$en + H_2O \rightleftharpoons enH^+ + OH^-$$
$$enH^+ + H_2O \rightleftharpoons enH_2^{2+} + OH^-$$

When two ligand donor atoms are brought into proximity upon bonding to a metal ion, an electrostatic repulsion between the negative charges or dipoles is experienced. In the case of two monodentate ligands, this interaction increases as ligands are brought together whereas in the case of a bidentate ligand the chelate effect is predominant. This can be explained by comparing the coordination of ammonia and ethylenediamine(en), in the case of NH_3 there is a greater unfavorable nitrogen-nitrogen repulsion involved in bringing the ligands together[1].

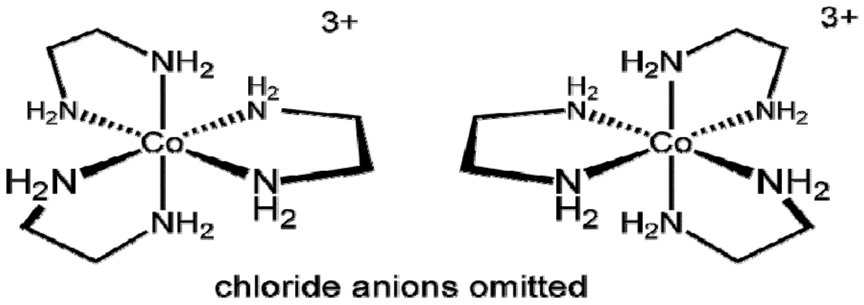


Scheme-11: Coordination Mode of Ammonia and Ethylenediamine(en).



Scheme- 12: Coordination Mode of a Typical Bidentate Ligand

In forming the chelate complex, as shown above in Scheme-12, there is a high probability of the second donor atom Y forming a bond to the metal whereas, with monodentate ligands, the probability is much lower. In other words, once the first M-L bond is formed the second donor atom is held close to the position required for the formation of the second bond. Ethylenediamine forms a large number of complexes with many different metal ions. It usually acts as a bidentate ligand and forming a five-membered chelate ring makes the complexes more stable. Concerning with the stereochemistry of the complex, there are two principal ways of orienting the three rings around the metal ion said to be enantiomers, which can be assigned as **Λ**∂∂∂ and **Δ**∂∂∂[1,33].



Λ∂∂∂

Δ∂∂∂.

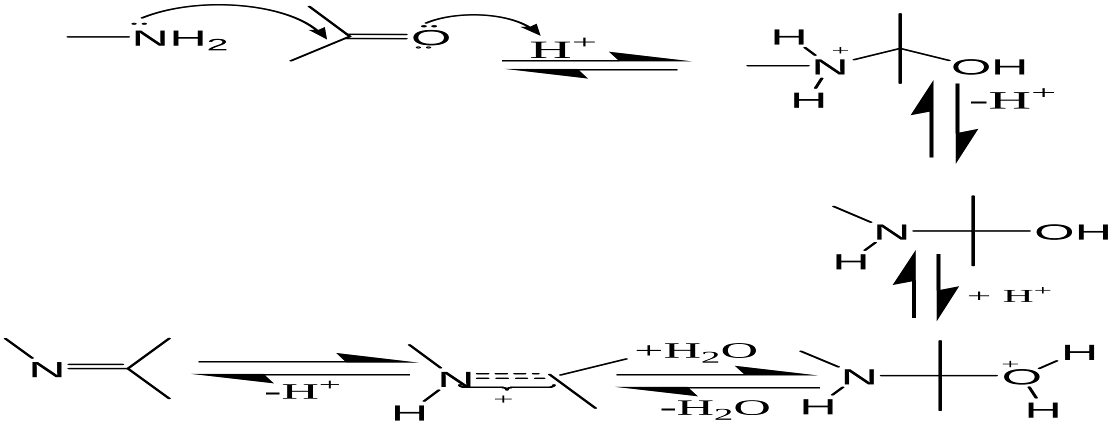
Figure-6: Orientation of the three rings around the metal ion in en complex.

In the former case (left-handed) the three rings are rotating to the left and the C-C bonds of the ring is parallel to the three-fold axis whereas in the later case (right-handed) the three rings are rotating to the right and the C-C bonds of the ring is perpendicular to the three-fold axis[1-3].

1.4. Schiff Base and Its Preparation

A Compound containing an imine or azomethine group(R-C=N-) is usually formed by the condensation of a primary amine with a carbonyl containing compound. The condensation of a primary amine with ancarbonyl compound provides an N-substituted imine known as Schiff's base after Hugo Schiff, a German chemist, who first reported the formation in 1864[16,37-40]. Schiff base preparation occurs in the two-step reaction that begins with nucleophilic addition of an aldehyde or a ketone with a primary amine, compound of the type RNH_2 or $ArNH_2$. In the first stage of the reaction the amine adds to the carbonyl group to give a species known as a carbinolamine. Once formed, the carbinolamine undergoes dehydration to yield the product of the reaction, an N-alkyl- or N-aryl-substituted imine. A Schiff base which is effective as coordinating ligand has a functional group such as OH, NH_2, SH etc.

There are several reaction path ways to the synthesis of a Schiff base. The most common one is an acid catalyzed condensation of an amine and a carbonyl compound as shown in (Scheme-13) [2-4,14,16] .

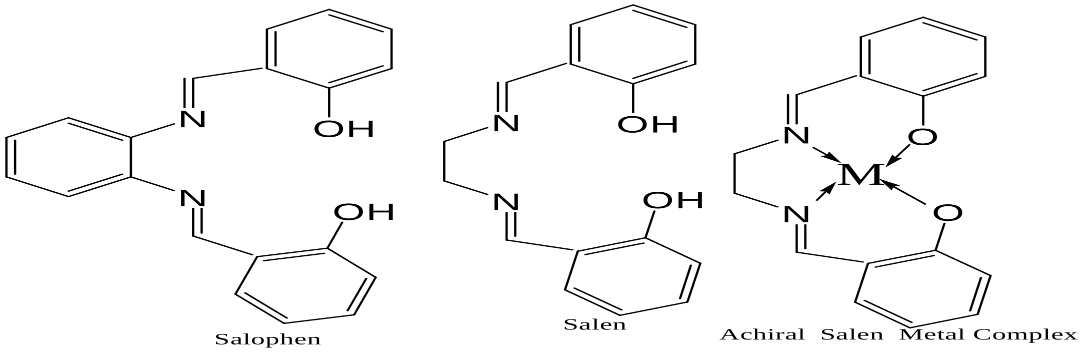


Scheme-13: Acid catalyzed Schiff base formation reaction

Common diamine Schiff base ligands can be prepared by condensation of salicylaldehyde or salicylaldehyde derivatives with 1,2-diamines lead to the formation of one extremely important class of ligands, generally known as

Salens, which are formed by combination of two equivalents of salicylaldehyde with a diamine (1,2-diamine) .

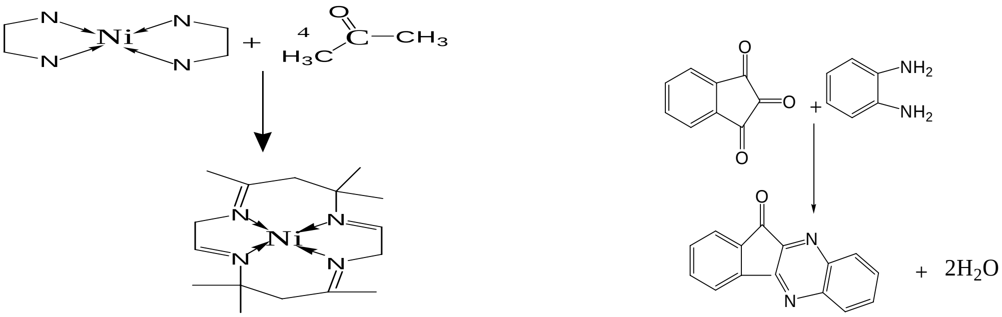
Although the term salen was used originally only to describe the tetradentate Schiff bases derived from ethylenediamine, the more general term Salen type is used to describe the class of [ONNO] tetradentate bis-Schiff base ligands (Figure-7). Metal complexes of salen schiff bases possess considerable potential in many applications including catalytic ,biological, clinical, analytical, microbial, insecticidial, antibiotics, tumor inhibitors [18-23] .



Salophen Salen Achiral Salen Metal Complex

Figure-7: Salen ligands and their metal complexes

Numerous researchers have attempted to synthesize a number of metal complexes of schiff bases from different carbonyl and amine containing compounds. For example the diamine complexes of Ni(II) and Cu(II) can under go further condensation reaction with acetone to give a poriphrin analogue (scheme-14), even the macrocyclic ligand can be synthesized in the absence of the metal ion[2,3,24] .



scheme-14: Schiff bases of diamines

The present project is proposed to develop a new schiff base from the condensation between ethylenediamine and ninhydrin.

1.5. The Chemistry Nickel (II), d^8

Ni (II) forms a large number of complexes the main structural types being octahedral, tetrahedral, and square-planar.

I. Octahedral complexes:

Octahedral Ni(II) complexes having $^3A_{2g}$ ground state are expected to have three spin-allowed transitions. $^3A_{2g} \rightarrow {}^3T_{2g}$, $^3A_{2g} \rightarrow {}^3T_{1g}$ (F) and $^3A_{2g} \rightarrow {}^3T_{1g}$ (P) in the ranges of 7000 - 13000, 11000 - 20000 and 19000 - 27000 cm^{-1} respectively. Two spin forbidden transitions are also possible, $^3A_{2g} \rightarrow {}^1E_g$ and $^3A_{2g} \rightarrow {}^1T_{2g}$. Magnetically, octahedral Ni(II) complexes have a relatively simple behavior. They all should have two unpaired electrons and thus posses magnetic moments ranging from 2.9-3.4 B.M. depending on the orbital angular momentum contribution.

II. Tetrahedral complexes:

For regular or nearly regular tetrahedral complexes there are characteristic spectral and magnetic properties. Naturally the more the irregular geometry of a paramagnetic Ni(II) complex, the less likely it is to conform to these specifications. The tetrahedral Ni(II) complexes with $^3T_1$ (F) ground state generally exhibit four transitions $^3T_1 \rightarrow {}^3A_2$, $^3T_1 \rightarrow {}^1E$, $^3T_1 \rightarrow {}^3T_1$ (P) and $^3T_2 \rightarrow {}^1T_1$.The band $^3T_1 \rightarrow {}^3T_1$ (P) is a strong band of high intensity when compared with others. The transition from the ground $^3T_1$ (F) state to the $^3T1$ (P) state occurs in the visible region (15,000 cm^{-1}) and is relatively strong ($\varepsilon \approx 102$) compared to the corresponding $^3A_{2g} \rightarrow {}^3T_{1g}$ transition in octahedral complexes. Thus tetrahedral complexes are generally strongly colored and tend to be blue or green unless the ligands also have absorption bands in the visible region . Because the ground state $^3T1$ (F) has much inherent orbital angular momentum, the magnetic moments of truly tetrahedral Ni(II) should be about 4.2 B.M. at room temperature. However, even slight distortions reduces this markedly (by splitting the orbital degeneracy).

Thus fairly regular tetrahedral complexes have magnetic moments of 3.5 - 4.0 B.M.; for the more distorted ones the magnetic moments are 3.0 - 3.5 B.M. (i.e., in the same range as six coordinated complexes).

III. Square planar complexes:

For the vast majority of four coordinated Ni(II) complexes, square planar geometry is preferred. This is a natural consequence of the d^8 configuration, since the square planar ligand set causes one of the d-orbitals ($d_{x^2-y^2}$) to be uniquely high in energy and the eight electrons can occupy the other four d-orbitals but leave this strongly antibonding one vacant. In tetrahedral coordination, on the other hand, occupation of antibonding orbitals is unavoidable. Square Planar complexes of Ni(II) are thus invariably diamagnetic. They are frequently red, yellow or brown owing to the presence of medium intensity absorption band ($\varepsilon \approx 60$) in the range 450 – 650 nm, but other colors do occur when additional absorption bands are present. Square planar Ni(II) complexes don't have any absorption band below 10000 cm^{-1}, due to large crystal field splitting. Hence they can be clearly distinguished from octahedral and tetrahedral complexes [1-4, 13,33,39] .

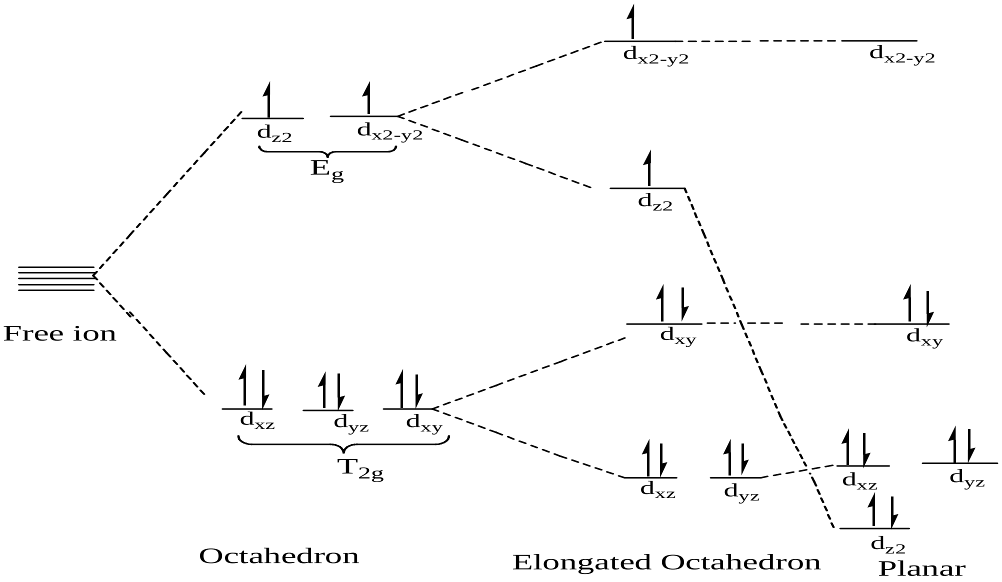


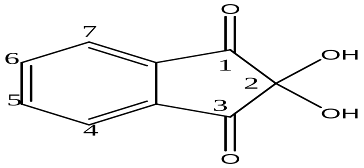
Figure-8 Schematic Splitting of d-orbitals in Ni(II)

1.6. Literature Survey

In relation to the biologically active and structurally significant Schiff base ligands and their metal complexes numerous Literatures have been developed**.**

The Chemistry of Ninhydrin

Ninhydrin has been used for the detection of amino acids and amines for almost fifty years, and many suggestions have been made as to the numbering system of the skeleton, the naming system, the sysnthesis and the mechanism of its reactions. Ninhydrin can named as triketohydrindene hydrate or Ruhemann's reagent or more systematically as 2,2-dihydroxy-1,3-indandione[7] .



Studies on the synthesis, characterization and antimicrobial activity of new Co (II), Ni (II) and Zn ((II) complex of Schiff base derived from ninhydrin and glycine

For the first time, Co(II), Ni(II) and Zn(II) complexes were synthesized involving an intermediate Schiff base, indane-1,3-dione-2-imine-N-acetic acid the condensed product of ninhydrin and glycine. These colored complexes were characterized by elemental analysis, molar conductivity, thermo gravimetric analyses/differential thermal analysis, infrared, magnetic susceptibility, NMR and electronic spectral studies. Mechanisms for their formation was proposed. The experimental studies revealed that the complexes possessed octahedral stereochemistry wherein the Schiff base behaved as a monobasic tridentate ligand. A molecular structure for the metal complexes was also proposed. A comparative study of the antimicrobial activity of ninhydrin and the corresponding metal complexes against Escherichia coli, Proteus mirabilis, Staphylococcus aureus and Streptococcus faecalis was undertaken and the results were discussed *[23]*.

Quantitative Reaction Cascades of Ninhydrin in the Solid State

Crystalline ninhydrin undergoes waste-free solid-state cascade reactions with dimedone, O-phenylenediamines, O-mercaptoaniline, ureas and thioureas, (Scheme-5,6&9). The yields are quantitative and give pure crystalline products without workup just by milling stoichiometric mixtures of the crystalline reagents. The structures of the new and the previously obtained products with lower yields from solutions are established or confirmed by spectroscopic data and density functional calculations. The waste-free and easy syntheses of highly functionalized (C-O; O-H; C-N) heterocycles or of a tetraketone are also of interest for synthetic use*[24]*.

The development of novel ninhydrin analogues

The first three ninhydrin analogues specifically synthesized for fingerprint development were reported[12]. Modification of the aromatic ring could favourably alter the optical properties of ninhydrin while preserving the reactivity of the cyclic triketone. The introduction of various substituents and extension of the conjugated system caused an increase in molar extinction coefficients yielding more sensitive reagents and induced shifts in the absorption maxima resulting in some variation in the colour of the Ruhemann's purple product[12].

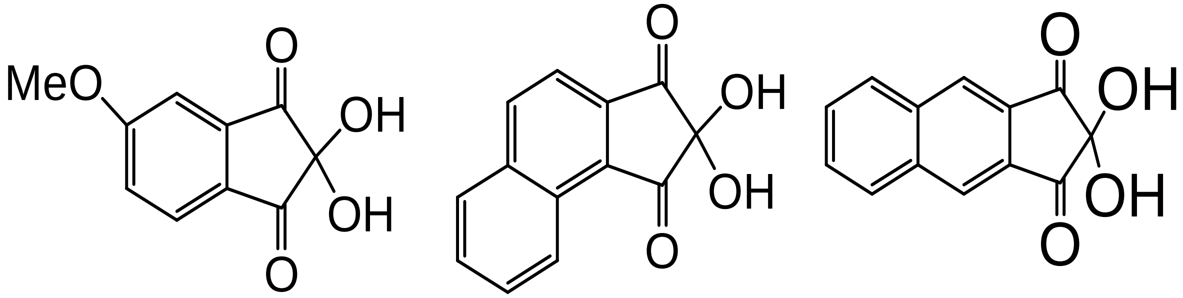
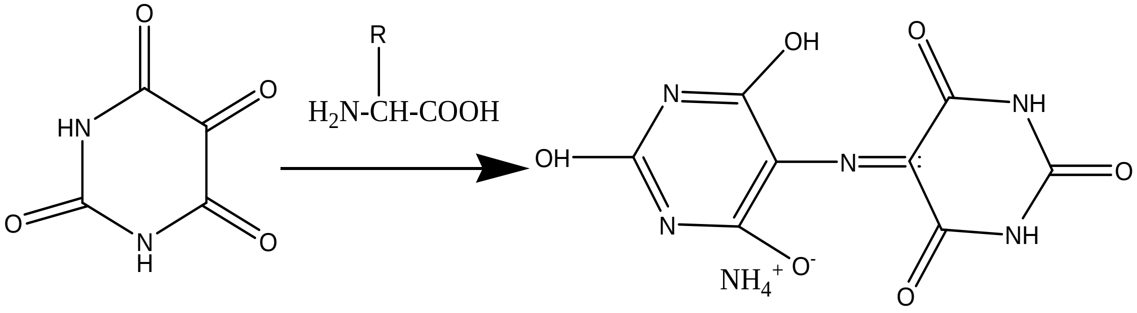


Figure-9: The first Ninhydrin Analogues

Due to structural similarities with ninhydrin and other cyclic triketones, like alloxan, has ability to react with amino acids to yield a blue compound(Scheme-15) [12].



Scheme-15: Reaction of Alloxan with Amino Acids

Synthesis and Characterization of Metal Complexes of Cu(II), Ni(II), Zn(II), Co(II), Mn(II) and Cd(II) with Tetradentate Schiff Bases

Two tetradentate ligands, (H_2L) and (H_2L') were formed by the 2:1 molar condensation of 4-(benzeneazo)salicylaldehyde with o-phenylenediamine or ethylenediamine. Their Cu(II), Ni(II), Zn(II), Co(II), Mn(II) and Cd(II) complexes were synthesized and characterized by elemental analysis, molar conductivity measurements, and infrared and electronic spectral data. Two ligands and their Ni(II) and Zn(II) complexes were further identified using ^{1}HNMR spectra. The results suggest that the metal is bonded to the ligand through the phenolic oxygen and the imino nitrogen[25].

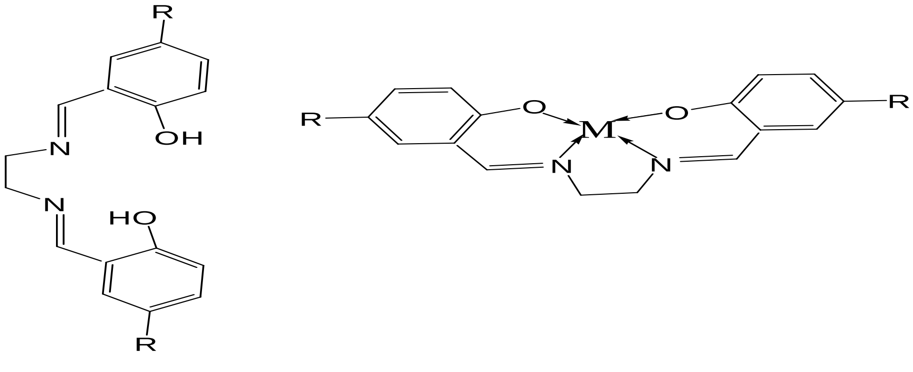


Figure-10: Schiff base ligand and its metal complexes (M=Cu, Ni, Zn, Mn or Cd)

The Direct Electrochemical Synthesis of Ti(II), Fe(II), Cd(II), Sn(II) and Pb(II) Complexes with N,N'-Bis(Salicylidene)-o-Phenylenediamine

The electrochemical oxidation of anodic (M = Ti, Fe, Cd, Sn, and Pb) into acetonitrile solutions of N,N′-bis(salicylidene)-o-phenylenediamine [SalophH_2] gives the corresponding M(Saloph) complexes in high yield. The mechanism of the electrochemical reactions is discussed. SalophH_2 forms complexes (1:1 molar ratio) with Ti, Fe, Cd, Sn, and Pb ions. The complexes have been characterized by elemental analyses, molar conductivity measurements, and infrared and electronic spectral data. The SalophH_2 complexes of Fe(II) and Cd(II) have been further identified by ^{1}HNMR and mass Spectra[26] .

Preparation, Physical Characterization and Antibacterial Activity of Ni (II)Schiff Base Complex

The Ni (II) Schiff base complex derived from salicylaldehyde and o-amino benzoic acid has been prepared and characterized. Several physical tools, in particular; elemental analysis, molar conductance, magnetic moment, infrared and electronic spectras were used to investigate the chemical structure of the prepared complex. The elemental analysis data show the formation of 1:1 [M:L] complex of the formula [NiLOH]H_2O, where L is the Schiff base compound under investigation. The molar conductance measurement reveals the presence of a non-electrolytic nature. The infrared spectral data of the complex display the complexation behavior of the Schiff base towards Ni (II) ion. The electronic spectrum of the complex exhibits the existence of $\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$(>C=N) transitions and the square planar geometry. The obtained magnetic moment value reveals a diamagnetic character. The free Schiff base and its complex have been tested for their antibacterial activities against several human pathogenic bacteria, and they appear to be promising as antibacterial agent against some specific bacteria[35] .

Synthesis, characterization and antiproliferative activity of metal complexes with the Schiff base derived from the 1:2 condensation of 2,6-diformyl-4- methylphenol and 5,6-diamino-1,3-dimethyluracil

A series of mononuclear complexes with Co(II), Ni(II), Cu(II), Zn(II), Hg(II), Mo(VI) and Pd(II) containing the ligand derived from the 1:2 condensation of 2,6-diformyl-4-methylphenol and 5,6-diamino-1,3-dimethyluracil (here after denoted as BDFDAAU) were synthesized. The complexes were characterized by elemental analysis, TG, ^{1}HNMR, ^{13}CNMR, UV–vis, IR and magnetic measurements. The deprotonated ligand in the phenolic oxygen shows a symmetric tridentate coordination mode through the two azomethine nitrogen atoms and the phenolic oxygen atom whereas the coordination of the neutral ligand takes place through the phenolic oxygen atom and one azomethine nitrogen atom. In the Mo(VI) complex, the ligand is bideprotonated in the phenolic oxygen and an amino group from one uracil unit; so, the coordination mode changes again into an asymmetric way: phenolic oxygen atom, one azomethine nitrogen atom and the nitrogen atom from the deprotonated amino group. The antiproliferative behaviour against the five human tumor cell lines (human neuroblastoma NB69, human breast cancer MCF-7 and EVSA-T, human glioma H4 and human bladder carcinoma cell line ECV) suggested a modulator behaviour, according to the concentration, of cell growth due to their estrogen-like characteristics[27] .

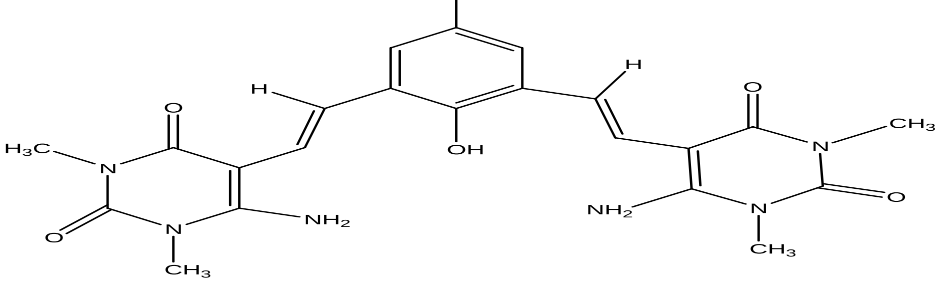


Figure-11 : The ligand BDFDAAU

Spectroscopic Characterization of Some Tetradentate Schiff Bases and Their Complexes with Ni(II), Cu(II) and Zn(II)

Three ligands have been formed by the 1:2 condensation of o-phenylenediamine with salicyldhyde,2-hydroxy-1-naphthaldhyde or o-hydroxyacetophenone. The potentially tetradentate ligands are N,N-bis(salicyldhyde)-o-phenylenediamine (SalophH$_2$), N,N-bis(2-hydroxy-1-naphthaldhyde)-o-phenylenediamine (NophH$_2$) and N,N-bis(o-hydroxyacetophenone)-o-phenylenediamine (AophH$_2$), respectively. These ligands form complexes (1:1 molar ratio) with in Ni(II), Cu(II) and Zn(II) ions. The complexes have been characterized by IR, ^{1}HNMR, MS, uv/vis spectra in addition to elemental analysis. The spectral data of the ligands and their complexes with Ni, Cu and Zn are discussed in connection with the structural changes which occur due to complexation[28] .

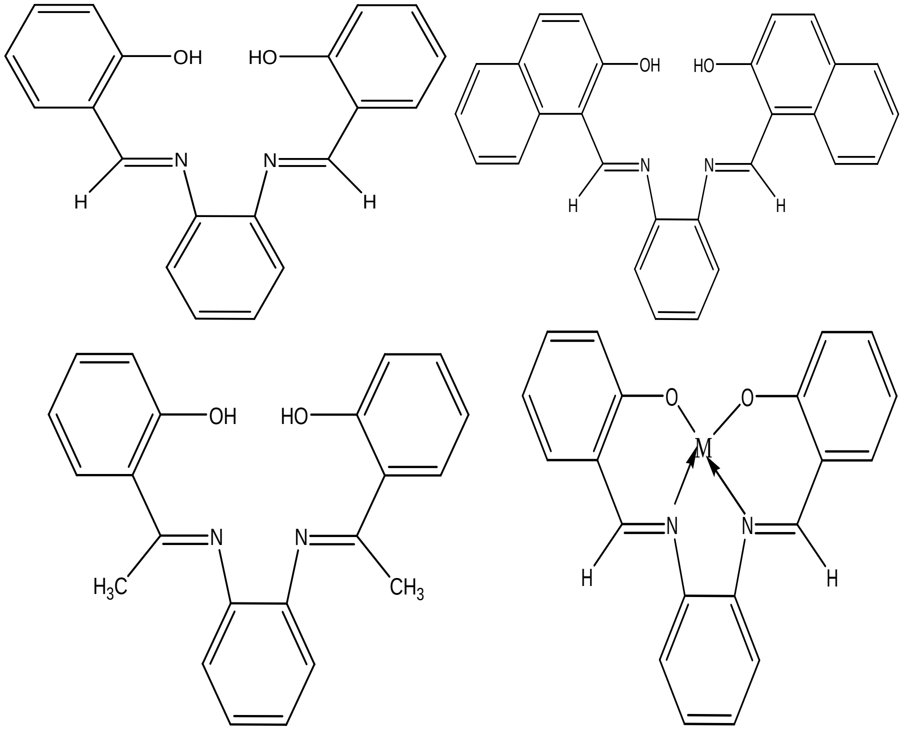


Figure-12 : O-phenylenediamine Schiff base ligands and thier metal complexes (M=Ni, Cu or Zn).

Synthesis, characterization and antifungal activity of a series of Mn (II) and Cu(II) complexes with ligands derived from reduced N,N'-O-phenylenebis(salicylideneimine)

A series of Mn(II) and Cu(II) complexes with reduced Schiff bases derived from o-phenylenediamine has been prepared and characterized by elemental analysis, TG measurements, ESR, magnetic measurements, FTIR, UV–Visible spectra and conductivity. These complexes were found to be [MnL(H_2O)$_n$] and [CuL](H_2O)$_n$ species with n = 0–2. Their antifungal activity was evaluated on different human fungi including yeasts of the Candida genus (C. albicans, C. glabrata, C. tropicalis and C. parapsilopsis) some opportunistic moulds belonging to the Aspergillus (A. fumigatus, A. terreus and A. flavus), Scedosporium genus (S. apiospermum and S. prolificans) and some dermatophytes (M. gypseum, M. persicolor, T. mentagrophytes, M. canis and T. tonsurans). The manganese complexes showed a significant growth inhibition of the dermatophytes tested and fungi of the genus Scedosporium. This is very interesting as these fungi are usually poorly susceptible to current antifungal including Amphotericin B and Itraconazole chosen as reference in this study[29] .

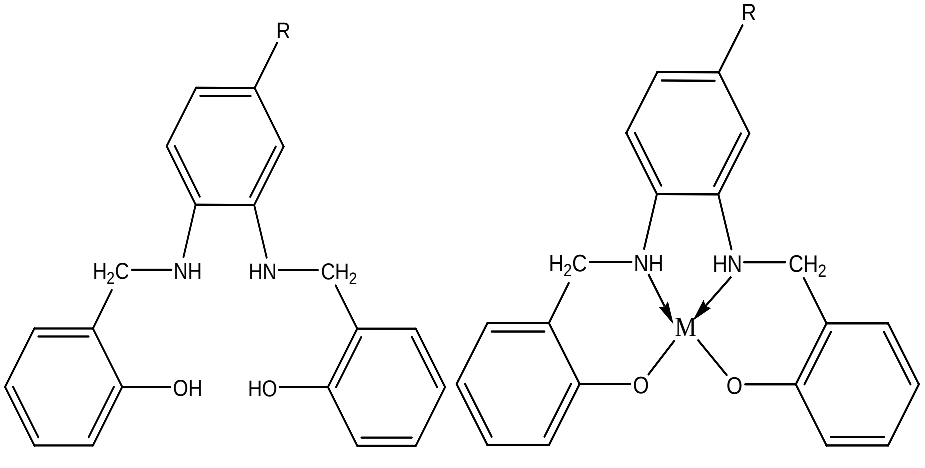


Figure-13 : O-phenylenediamine reduced Schiff base ligand and its metal complexes(M=Cu and Mn)

1.7. Objective of The Present Investigation

A search through literature reveals that a lot of work has been done on different metal complexes of Schiff bases derived from ninhydrin and α-amino acids or amines, which have ON donor sites. However no systematic work is reported on metal complexes of ninhydrin–aliphatic diamine derivative. Thus the present study aimed to synthesize and characterize a new ligand through the derivatization of ninhydrin and ethylenediamine and investigate the reaction of the product with a selected transition metal ion.

In view of the wide range of potential applications of ninhydrin derivatives and their metal complexes in agriculture, pharmaceutical and analytical field, it has been aimed to synthesize and study such ligand and its Ni (II) complex. In this project the synthesis and characterization of the ligand and its metal complex containing Ni(II) will be presented. The reaction between ninhydrin and ethylenediamine is likely to result in product which may be of 1:1, 1:2, or 2:1 mole ratio with respect to the reactant. But any of these products can be a potential chelating system towards transition metal ion. Studying the coordinating property of the ligand towards Ni(II) ion using IR and UV-vis spectra, atomic absorption spectroscopy, conductivity measurements and magnetic studies is the objective of this work. The objectives set below will be attempted during the investigation:

A) Synthesis of the Schiff base ligand from ninhydrin and ethylenediamine .

B) Synthesis of its Ni(II) complex.

C) Characterization of the synthesized ligand and Ni(II) complex using analytical,

spectral, conductance and magnetic studies.

2. EXPERIMENTAL

2.1. Materials and Methods

2.1.1. Chemicals, Reagents and Solvents

All chemicals, reagents and solvents used were of Analar grade, conc.HNO_3, $NiCl_2.6H_2O$, $AgNO_3$ (0.1M), dimethylglyoxime ($C_4H_8O_2N_2$), ninhydrin, ethylenediamine, methanol, ethanol, acetone, dichloromethane, DMSO ,hexane, aqueous ammonia, acetonitrile and chloroform.

2.1.2. Instrumentation

Melting/decomposition temperatures were determined with SMP3, Digital Melting point apparatus. The purity of the complex was checked by thin layer chromatography(TLC). Elemental Analysis of the Ligand was measured by Instrument-CE-440 Elemental Analyzer, University of Nottingham, School of Chemistry UK. Molar conductivity of the complex was measured at room temperature with freshly prepared solution in ethanol using a JENWAY 4330 conductivity and pH meter. Magnetic susceptibility measurement of the complex was recorded at 26°C on a MSB-AUTO Magnetic balance (Sherwood Scientific). The metal content was estimated by using BUCK SCIENTIFIC ATOMIC ABSORPTION SPECTROPHOTMETER MODEL 210 VGB. Electronic spectra of the ligand and complex were recorded using SPECTRONIC GENESYS 2PC Uv-Visible spectrometer with a 1cm cell in acetonitril and ethanol respectively at room temperature.

Infrared spectra (KBr pellet) were recorded on Perkin Elmer Spectrum BX FT-IR spectrometer in the range of 4000-400 cm^{-1}.

2. 1.3. Methods

A. Qualitative Tests

I.Thin Layer Chromatography (TLC)

TLC was used to check the purity of the compounds. For this purpose 2x4 cm silica coated aluminum plates were used and a suitable solvent or mixtures of solvents in certain propositions were used as mobile phase. Observation of a single spot indicates the purity of the compound.

II. Chloride Test

The metal complex was digested in nitric acid and was subjected to chloride identification. A white precipitate formed in the solution after addition of $AgNO_3$ solution indicates the presence of chloride ion in the complex.

III. Metal Ion Test

The presence of Ni(II) ion in the complex was tested by using dimethylglyoxime. When the Ni(II) complex dissolved in nitric acid was treated with dimethylglyoxime solution, it forms a red precipitate, which confirms the presence of Ni(II) ion.

B. Quantitative Determinations

I. Metal Estimation

The metal content in the complex was determined using atomic absorption spectroscopy. The experimental percentage of metal in the complex was found as:

$$M\ (\%) = \text{Absorbance (A, ppm)} \times \frac{\text{Volume diluted to} \times 100}{\text{Mass of the sample taken} \times 1000}$$

The result obtained was compared with the theoretically expected value to obtain the metal to ligand ratio in the complex.

II. Molar Conductance Measurement

The molar conductance was determined by taking 10mg of the complex in 25ml of ethanol .

III. Magnetic Susceptibility Measurement

Many transition metal salts and complexes are paramagnetic due the partially filled d-orbitals. MSB Auto, Sherwood instrument can generate grame susceptibility (χg) data for a given paramagnetic substance. The following calculations were made to arrive at the magnetic moments. Molar magnetic susceptibility (χ_M) = χg X Molecular weight of the compound χ_M is subjected to diamagnetic correction using Pascal constants and to obtain the corrected magnetic Susceptibility (χM^{corr}), $\chi M^{corr} = \chi_M - \chi dia$

from which the magnetic moment is finally calculated from the equation :

$$\mu eff \text{ (effective magnetic moment)} = 2.824(\chi M^{Corr}.T)^{1/2}$$

$$\boldsymbol{\mu eff = 2.824(\chi M^{Corr}.T)^{1/2}}$$

2.2. Synthesis

2.2.1. Synthesis of The Ligand

The Ninhydrin, 2g (11.24 mmol) was dissolved in 30 ml ethanol, which gives a yellowish solution and ethylenediamine, 0.4 ml (5.62 mmol) was added to it by using a syringe while stirring. The mixture was stirred for 12 hours until a dark-brown precipitate was formed . The precipitate was filtered under sunction and repeatedly washed with ethanol. The product was then dried in open air and stored in a desiccator. Yield = 32.50%, Color = Dark-brown, Appearance = Fine powder, Empirical formula =H_2 L ($C_{20}H_{12}N_2O_4$.).

Elemental Analysis Data: %Calc (Found), C=69.8 (70.8), H=3.2 (2.9), N = 7.6 (7.7)

IR Data(cm^{-1}): v(OH) = 3450 – 3200, v(C=O)=1711, v(C=N) = 1618,
v(C=C) = 1582, v(CH) = 2924.

Uv-Vis. Data(cm^{-1}): π→π*(aromatic)=40,816, n →π*(C=O)=36,496 and
n →π*(C=N) = 23,981.

2.2.2. Synthesis of Ni(II) Complex

$NiCl_2.6H_2O$, 0.25 g (1.05 mmol) was dissolved in 30 ml warm ethanol and 0.40 g (1.05 mmol) of the ligand dissolved in 30 ml acetonitrile was added. The mixture was refluxed for 14 hrs and the brick-red precipitate formed was cooled and filtered under sunction and repeatedly washed with a mixture of ethanol and acetonitrile (50%). The product was then dried in open air and stored in a desiccator.

Yield=30.70%, Color=Brick-red, Appearance = Crystalline,

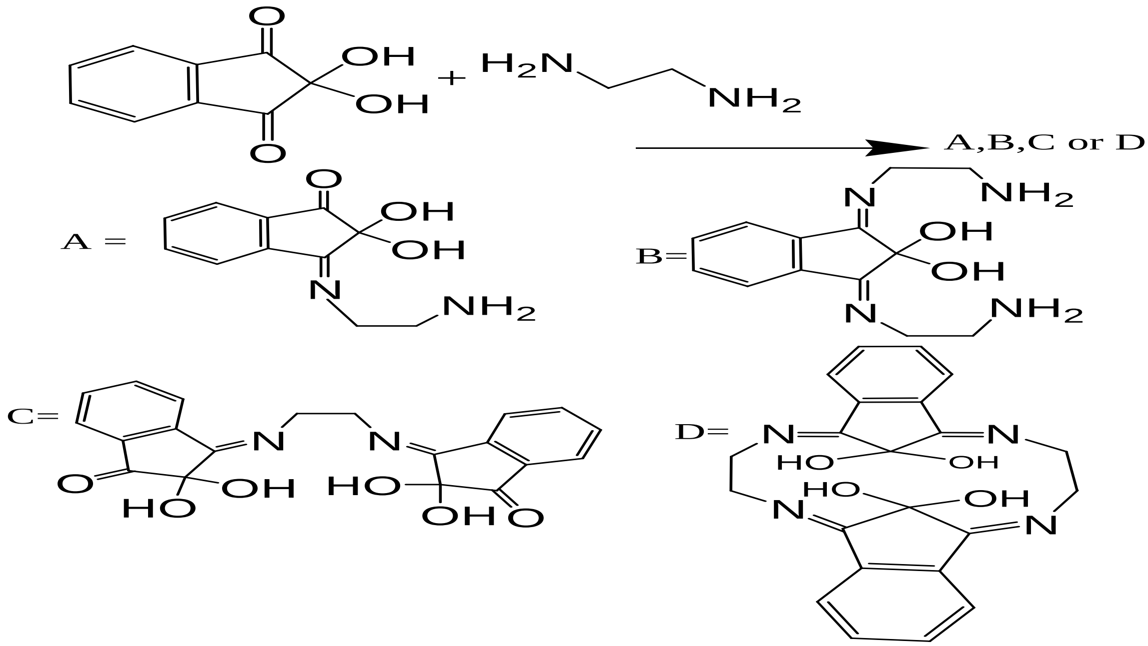
Λ_M =2.5 S cm^2 mol^{-1}, Empirical formula = $NiL.6H_2O(NiC_{20}H_{24}N_2O_{10})$.

IR Data(cm^{-1}): v(OH)=3500-30000, v(C=O)=1723, v(C=N)=1624, v(C=C)=1586, v(CH)=2924, υ(M-N)imin= 611, υ(M-O)=528

Uv-Vis. Data(cm^{-1}): Intraligand(28,490, 35,336 and 40,000) and
d-d transitions(10,526, 15,601 and 20,202).

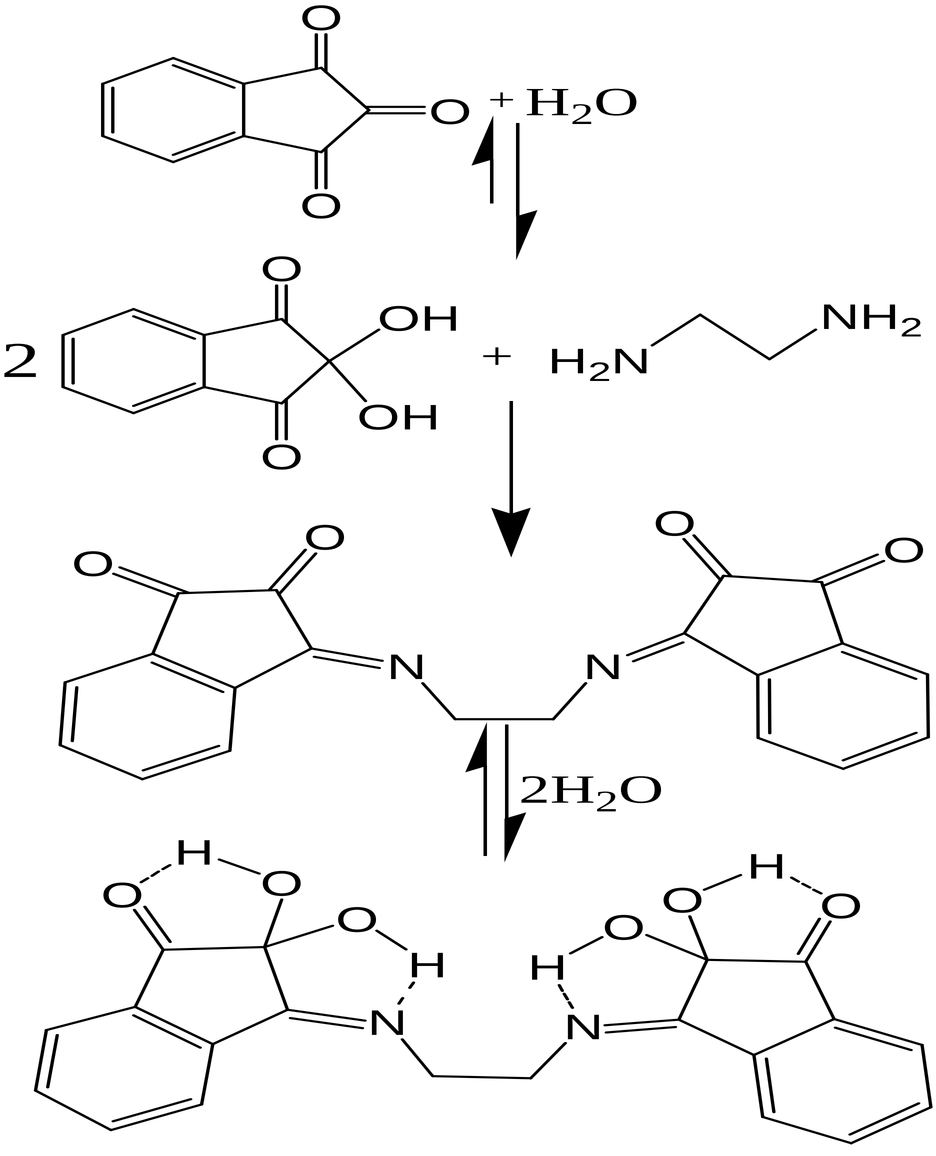
3. RESULTS AND DISCUSSION

Generally, condensation of ninhydrin with amino acids or primary amines give a deep purple compound known as Ruhemann's purple, maximally absorbs at 570 and 416 nm. The compound is believed to be formed via several steps; mainly condensation of the amine group with the carbonyl group of ninhydrin to give a ketimine Schiff base, decarboxylation and further condensation with another ninhydrin to give the final product (Scheme-8). In the present investigation; condensation of ninhydrin with ethylenediamine (2:1 molar ratio); it is shown that the reaction does not lead to the formation of Ruhemann's purple instead it gives a tetradentate ONNO donor Schiff base ligand (H_2L), which is coordinated with Ni(II) to form an octahedral complex. The reaction between ninhydrin and ethylenediamine is likely to result in product which may be of 1:1, 1:2, or 2:1 mole ratio with respect to the reactant(scheme-16), product A, B, C or D may be obtained. But any of these products can be a potential chelating system towards transition metal ion.



Scheme-16: Condensation of Ninhydrin with Ethylenediamine in various mole ratio.

Based on analytical (elemental analysis) and spectral data, from the above condensation products compound **C** is obtained. As the parent 1,2,3-trione tutomerizes with the 2,2-dihydroxy species (ninhydrin) and the ligand (H_2L) also expected to tutomerize with the 1,2-diketo.



H_2L

Scheme-17: Synthesis of the Ligand(H_2L)

Reaction of Ni(II) with the tetradentate Schiff base ligand (H_2L) in 1:1 molar ratio in a mixture of ethanol and acetonitrile (50%) solvent gives the metal complex, $NiL.6H_2O$ (Scheme-18).

$$NiCl_2.6H_2O + H_2L \xrightarrow[80^0c\text{ , }14hrs]{\text{ethanol+acetonitril(50\%)}} NiL.6H_2O$$

Scheme-18: Synthesis of the Complex

Some of the important physical characteristics of the synthesized compounds (ligand and complex) are listed below. The ligand has a dark-brown color while the complex is brick-red and insoluble in water and most organic solvents, while it is soluble in solvents with high dielectric strength like DMF, DMSO and acetonitrile (Table-1). As elemental analysis data indicates the analysis of carbon, hydrogen and nitrogen of the ligand matches adequately with the formula $\mathbf{C_{20}H_{12}N_2O_4}$, the data is presented in Table-2.

Table-1: Solubility of the ligand in different solvents

Compound	solvents	solubility
Ligand**(H_2L)**	Water	insoluble
	Methanol	Partially soluble
	Ethanol	Partially soluble
	Chloroform	Partially soluble
	DMSO	soluble
	DMF	soluble
	acetonitrile	soluble

Table -2: Analytical data and some physical properties of ligand & complex

Cpd	**Formula**	**M.Wt.**	**Color**	**Yield (%)**	**M.Pt (C^0)**	**Appearance**	**% Composition Calc.(found)**		
							C	H	N
H_2L	$C_{20}H_{12}N_2O_4$	344	Dark-brown	32.50	229-231	fine powder	69.8(70.8)	3.2 (2.9)	7.6(7.7)
Cpx	$NiL.6H_2O$	544.71	Brick-red	30.70	305-307	crystalline	----------	---------	-------

L=Ligand, Cpd=Compound, Cpx=Complex, M.Wt=Molecular Weight, M.Pt=Melting Point

3.1. Characterization of The Ligand

3.1.1. Infrared Spectrum (Band Frequency in Cm^{-1})

In its trione form ninhydrin shows three bands in the C=O stretching region : 1760, 1750 and 1720. The 1750 and 1720 bands are characteristic bands of its 1,3-dicarbonyl functional group and the 1760 band is characteristic of the intermediate one, which is in equilibrium with the 2,2-dihydroxy species. In the product, from the two C=O stretching bands (1748 and 1718) of the fresh 2,2- dihydroxy species the one with higher frequency (1748) is not observed, which indicates the derivatization of this group, a new strong and sharp band is observed at 1618 which is absent in free ninhydrin, assignable to the azomethine (N=C) group. Azomethine (N=C) group absorbs in the region 1690-1640 (v), but in aromatic ketimines, it absorbs in the lower region 1640-1620. Other supportive bands of the derivatization are the bands at 2924 and 2876 which are due to CH_2 asymmetric and symmetric stretchings respectively[11,21,30,34,36].

3.1.2. Electronic Spectrum

The electronic spectrum of the ligand was recorded at room temperature using acetonitrile as a solvent.. The spectrum of the Schiff base ligand consists of three main bands at 245 nm, 274 nm and 417 nm. The band at 245 nm can be assigned to the $\pi \rightarrow \pi^*$ transition of the aromatic group of nihydrin and the second band at 274 nm can be assigned to the $n \rightarrow \pi^*$ transition of the carbonyl group of nihydrin[11,21,32,33] .

3.2. Characterization of Ni (II) Complex

The interaction of hexahydrated Ni(II) metal chlorides with the ligand resulted in the formation of a Ni(II) complex. The complex is colored (brick-red), solid, air-stable, insoluble in water, soluble in warm ethanol and methanol, and also in DMSO and DMF.

3.2.1. Analytical Studies

The metal complex was initially tested for the presence of chloride and metal ions. For this purpose 10 mg of the complex was decomposed by HNO_3 and the qualitative test for chloride ion (with $AgNO_3$) and for Ni(II) ion (with dimethylglyoxime) were performed. The result confirms the absence of chloride ion and the presence of the metal in the complex.

3.2.2. Estimation of Nickel (by AAS)

Subsequent to qualitative testing, the metal estimation and metal to ligand ratio of the complex was obtained using atomic absorption spectroscopy and the result indicates the metal to ligand ratio to be 1:1.

3.2.3. Molar Conductivity

The molar conductance of the complex was recorded at room temperature by using ethanol and the value was found to be, Λ_M =2.5 S cm^2mol^{-1} indicates the non electrolyte nature of the complex.

3.2.4. Magnetic Susceptibility

There is a direct relationship between the magnetic properties of matter in the bulk and the number of unpaired electrons. The magnetic moment of Ni(II) complex was determined from the magnetic susceptibility measurement at 26 ^{0}C. The gram susceptibility of Ni(II) was found to be $3.5X10^{-6}$ and the magnetic moment calculated is 2.92 B.M. which indicates the presence of two unpaired electrons. The μeff value close to spin only value supports Octahedral stereochemistry. As such, the Ni(II) complex is proposed to be Octahedral in geometry[1-4,33].

Table-3: Molar conductivity and Magnetic moment data of the complex

Compound	Λ_M (S cm^2 mol^{-1})	Electrolyte Nature	Magnetic moment(B.M.)	
			Theoretical	Experimental
Ni-L	2.5 (<35)	non Electrolyte	2.9-3.2	2.92
	35-45	1:1 Electrolyte		
	70-90	2:1 Electrolyte		

3.2.5. Infrared Spectrum (Band Frequency In Cm^{-1})

The IR spectra show a band over 3500–3000 range assignable to *v*(O-H) vibration which may be broadened due to the presence of coordinated and lattice water molecules or by the establishment of an intramolecular H-bond. The comparison of the IR spectra of the complex and the ligand shows a displacement of the *v*(C=N) and *v*(C–O) vibration modes. The *v*(C=N) stretching vibration is displaced to the higher frequency, because Ni(II), d^8 is electron rich; in the coordination the azomethine (N=C) group may act as sigma donor and pi acceptor system to form (N→M). The *v*(C–O) vibration modes also shows a shift due to coordination. In addition the complex shows new bands: υ(M-N) imin= 611 and υ(M-O)= 528. Other supporting band is stretching vibration band of υ(M-O-C)= 1100(m). The complex shows one strong band in the region at 1723 which is characteristic of the one carbonyl functionality and $v_a(CH_2)$= 2924, $v_s(CH_2)$= 2846, δ(CH_2)= 1450 for the ethylene group, *v*(C-H) and *v*(C=C) of the aromatic group are also observed at 3065 and 1586 respectively [11,21,30-34,36].

Table-4 IR spectral data of the starting materials , ligand and complex (KBr pellets)

Abs. Bands	*vOH* *cm-1*	v_a*NH* v_s*NH* *cm-1*	v_a*CH* v_s*CH* *cm-1*	*vC=O* *cm-1*	*vC=N* *cm-1*	*vC=C* *cm-1*	*vC-O* *cm-1*	δ(O-H) In plan Out of plan	υ(M-N)imin υ(M-O) *cm-1*
En	------	*3368,* *3284*	*2924,* *2846*	-------	------	------	-----		---------
Nin.	*3400-3000*	------- -------	------	*1748,* *1718*	-------	*1582*	*1153*	*1521* *742*	---------
(L)	3450-3200	------- -------	2924, 2846	1711	1618	1582	1130	1535 733	--------
Ni-L	3500-3000	------- -------	2924, 2846	1723	1624	1586	1100	1534 731	611 528

3.2.6. Electronic Spectra of Ni(Ii) Complex

The electronic absorption spectra of the ligand and its Ni (II) complex were recorded at room temperature using the solvent acetonitrile and ethanol respectively. The Ni(II) complex shows six major bands at 250 nm, 283 nm, 351 nm, 495 nm, 641 nm and 950 nm. The features of the spectra are explained below. The band at 250 nm is attributed to $\pi \rightarrow \pi^*$ transition of the aromatic region of the ninhydrin and the band around 283 nm is due to the $n \rightarrow \pi^*$ transition of the C=O group. These two bands do not show any significant change. The band at 351 nm is attributed to $n \rightarrow \pi^*$ transition of the hydroxylate anion($C\text{-}O^-$) and the band around 495 nm is assigned to be $n \rightarrow \pi^*$ transition of the azomethine (N=C) chromophore, which shows significant change due to coordination.

The electronic spectrum of Ni-L complex shows characteristic bands in Visible/near IR region which are assignable to d-d transition. Ni-L complex has three spin allowed transitions ; $^3A_{2g} \rightarrow {}^3T_{2g}$, $^3A_{2g} \rightarrow {}^3T_{1g}(F)$ and $^3A_{2g} \rightarrow {}^3T_{2g}(P)$ at 950 nm (10526 cm^{-1}),

641 nm (15601cm^{-1}) and 495 nm (20202 cm^{-1}) respectively. The third band with the highest energy might over lap with the band of azomethine (N=C) chromophore. Based on the data presented in Table-5 along with assignment of transitions, octahedral geometry has been assigned to Ni–L complex [11,13,21,32,33,39].

Table-5 Electronic spectra of the Ligand and Ni(II) complex

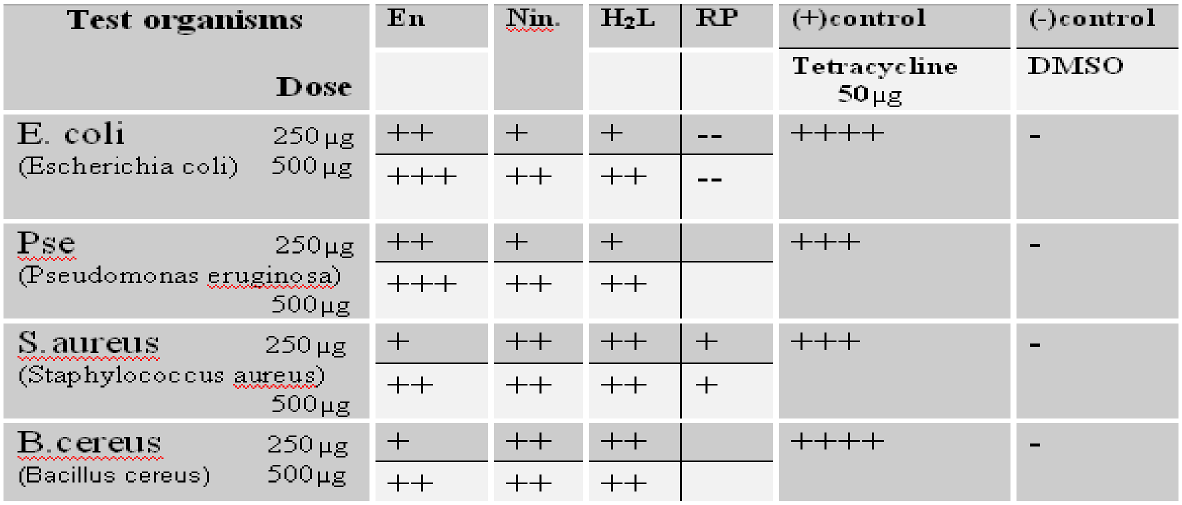
Compound	Assignment of Transition	Band Positions			
		Experimental		**Theoretical**	
		λmax/nm	υ/cm^{-1}	λmax/nm	υ/cm^{-1}
Ligand(H_2L)	$n \rightarrow \pi^*$(C=N)	417	23,981	400-430	25000-23256
	$n \rightarrow \pi^*$(C=O)	274	36,496	270-300	37,037-33,333
	$\pi \rightarrow \pi^*$(aromatic)	245	40,816	230-250	43,478-40,000
Ni(II)Complex	$^3A_{2g} \rightarrow {}^3T_{2g}$	950	10,526	1429-769	7,000-13,000
	$^3A_{2g} \rightarrow {}^3T_{1g}$(F)	641	15601	909-500	11,000-20,000
	$^3A_{2g} \rightarrow {}^3T_{1g}$(P)	495	20202	526-370	19,000-2,7000
	Intraligand	351,283, 250	28490,35336,40000	---------------	-----------------

3.3. ANTI-MICROBIAL STUDIES

The diameter of the inhibition zones around each sample, controls and solvents were measured. These data were then converted into a conventional way of expressing the bacterial activity (+ and -) by taking some approximate ranges and comparing them with the activity of the starting materials, ninhydrin and ethylenediamine.

Ethylenediamine was found to be highly active against the gram-negative bacteria than gram-positive bacteria. But the reverse was true for ninhydrin. The ligand showed an activity of in between the two (Synergetic effect). On the other hand Ruhemann's Purple had no activity on gram-negative bacteria but it showed some activity on gram-positive bacteria. In all cases the effect might be associated with structural difference. The activities of the synthesized ligand, ninhydrin and ethylenediamine against the four test organisms and Ruhemann's Purple for two test organisms are given in Table-6[14]. Many of the compounds were found to be active against the two gram-negative bacteria (Escherichia coli and Pseudomonas eruginosa) and the two gram positive bacteria (Staphylococcus aureus and Bacillus cereus). The inhibiting activity of the derivatized compound, the ligand; against the gram negative and gram positive bacteria was found to be in between of the starting materials (ninhydrin and ethylenediamine). These effects may be associated with the structure of the compounds[14,27,29,35].

Table-6: Antimicrobial test result



Test organisms	Dose	En	Nin.	H_2L	RP	(+)control	(-)control
						Tetracycline 50 µg	DMSO
E. coli (Escherichia coli)	250 µg	++	+	+	--	++++	-
	500 µg	+++	++	++	--		
Pse (Pseudomonas eruginosa)	250µg	++	+	+		+++	-
	500µg	+++	++	++			
S. aureus (Staphylococcus aureus)	250 µg	+	++	++	+	+++	-
	500µg	++	++	++	+		
B.cereus (Bacillus cereus)	250 µg	+	++	++		++++	-
	500µg	++	++	++			

En = Ethylenediamine, Nin. = Ninhydrin, H_2L = Ligand, RP = Ruhemann's Purple

Key. **- =no activity, v+ = 5-10 mm (some activity),**

++ = 10-15mm (moderate activity),

+++ = 16-20mm (high activity),

++++ = 21-25mm (very high activity).

4.CONCLUSION

Ninhydrin and ethylenediamine undergo a condensation reaction to give a Schiff base Ligand which forms distinctly colored complex with Ni(II). The Schiff base ligand was shown to behave as a dibasic tetradentate ONNO donor system and form stable chelate along with two moles of water in the coordination sphere. On the basis of analytical, conductance, IR spectra , electronic spectra and magnetic susceptibility data the geometry of the complex has been concluded to be octahedral(Figure-14).

The bacterial activity of the starting materials (ninhydrin and ethylenediamine), the ligand and Ruhemann's Purple was tested and all of the compounds showed activity against the tested gram-negative and gram-positive bacteria . But due to time frame the bacterial activity of the metal complex was not done, so further investigations are necessary for any possible anti-microbial or medicinal applications.

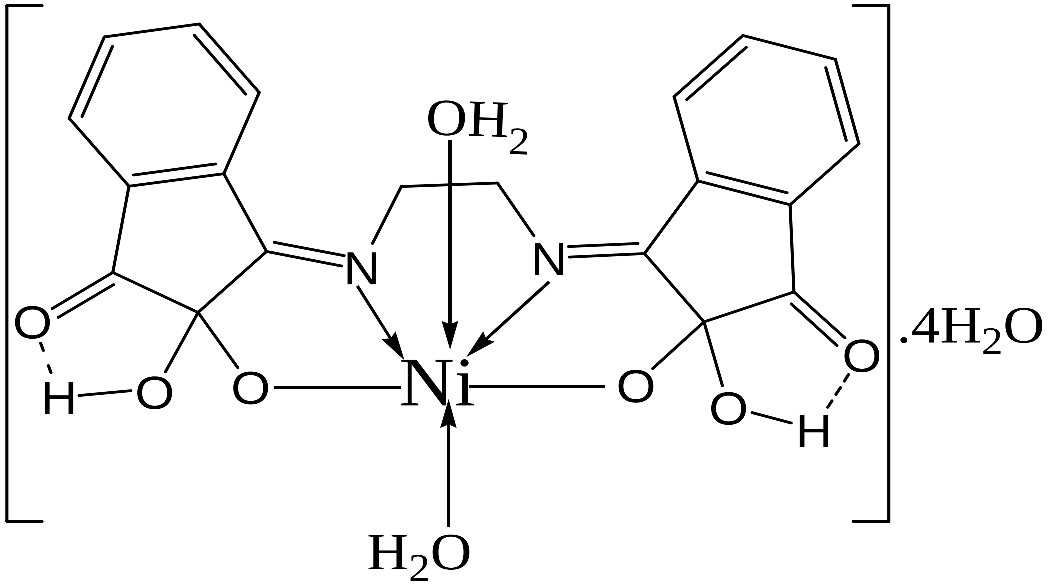


Figure- 14: Proposed structure of the Complex

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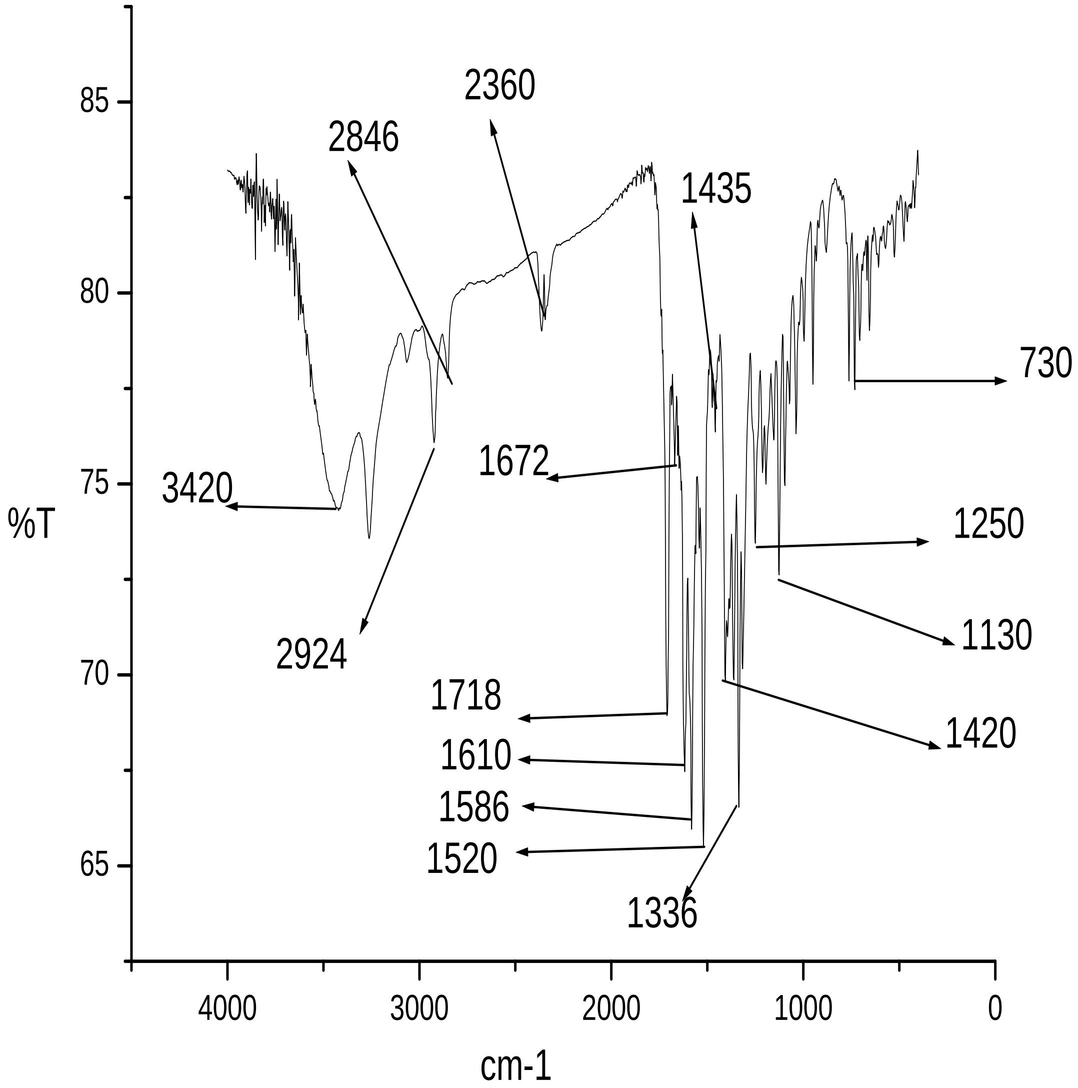
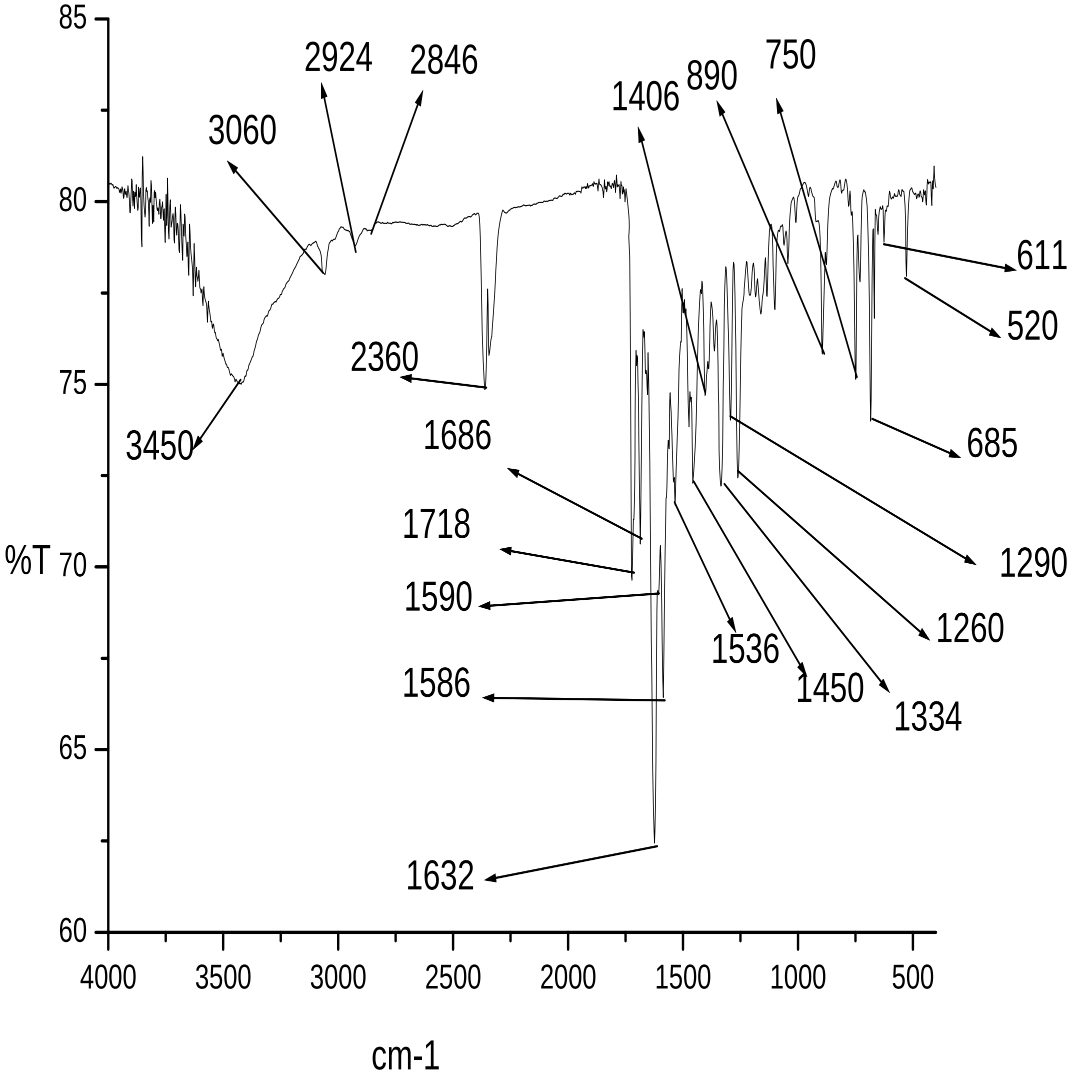
List of Appendices

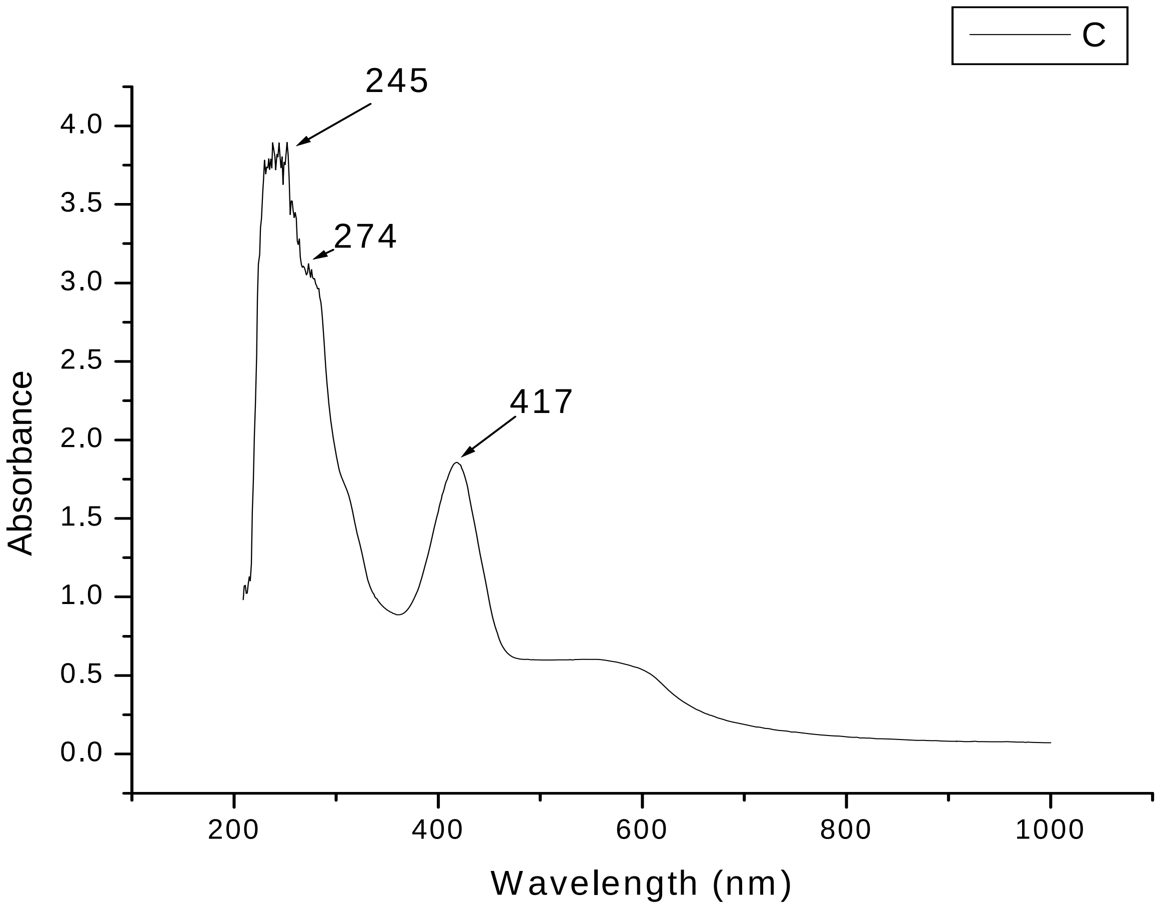
Appendix IR Spectrum of The Ligand



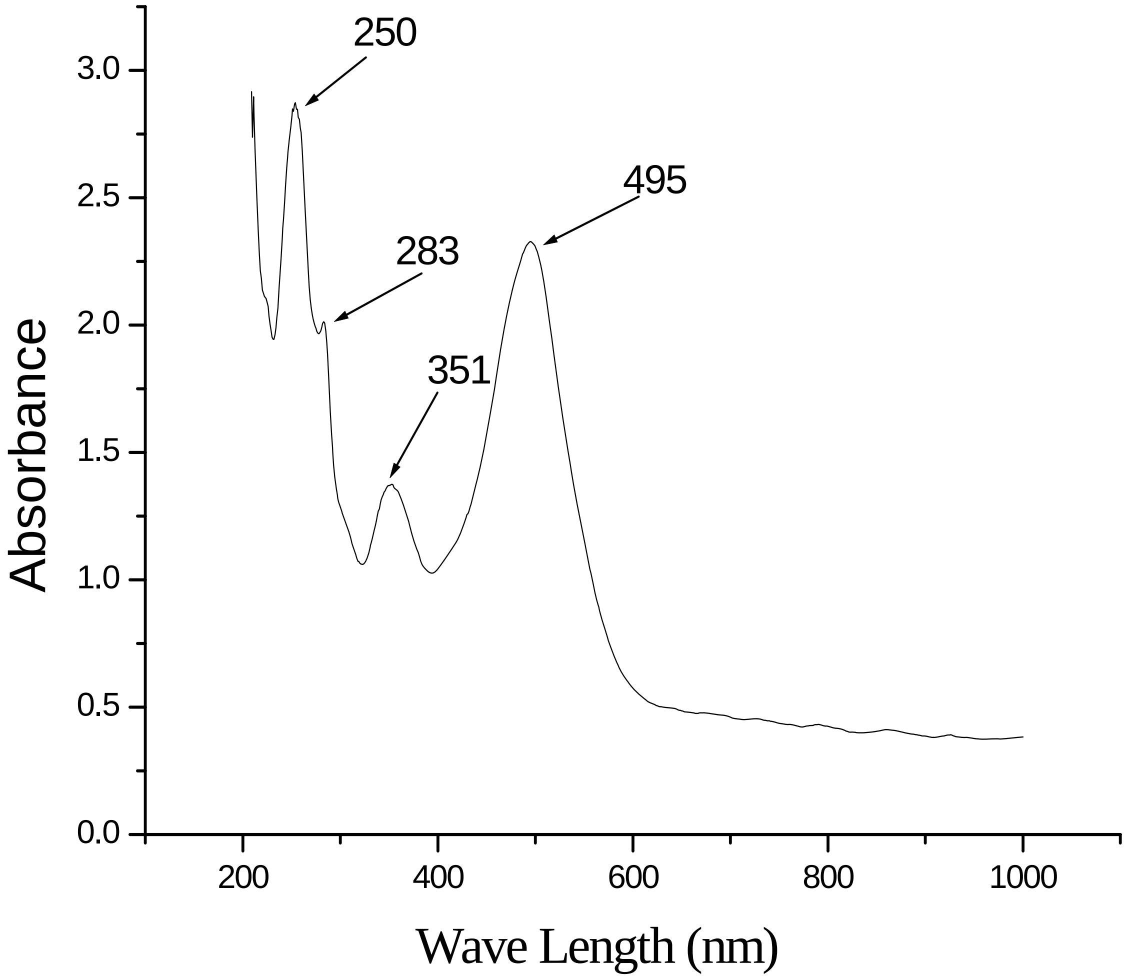
Appendix IR Spectrum of The Complex



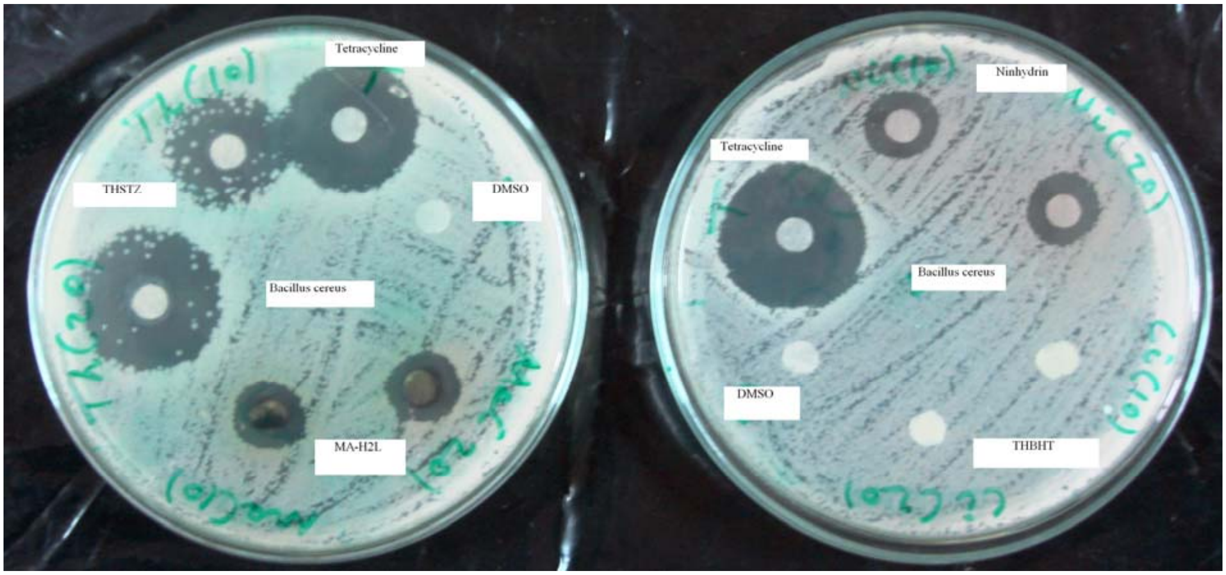
Appendix UV-Vis Spectrum of The Ligand

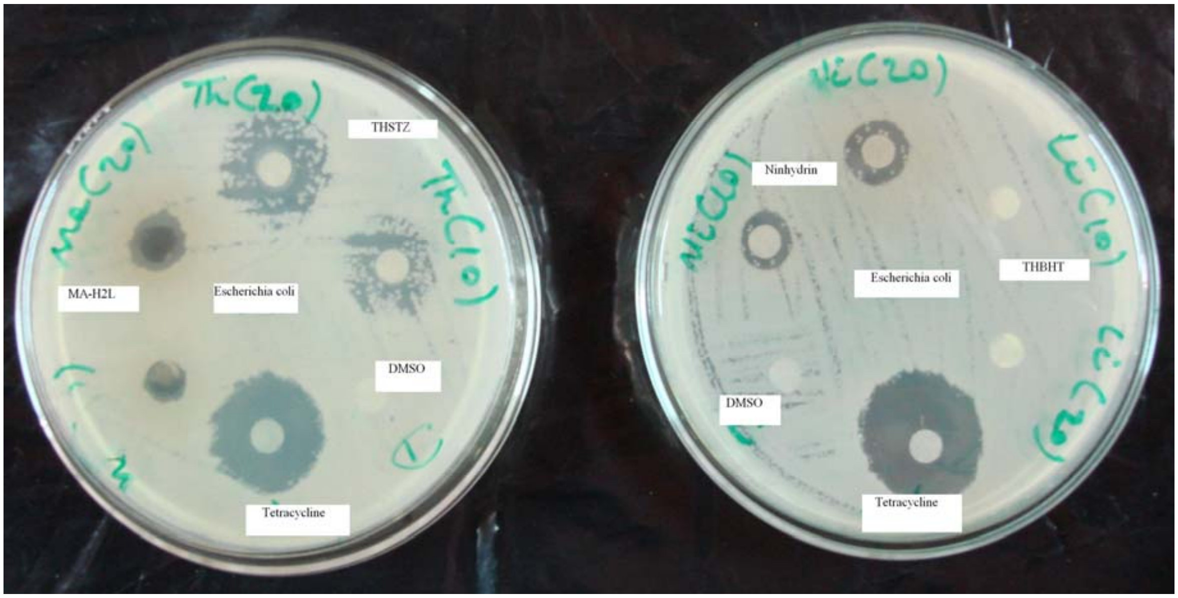


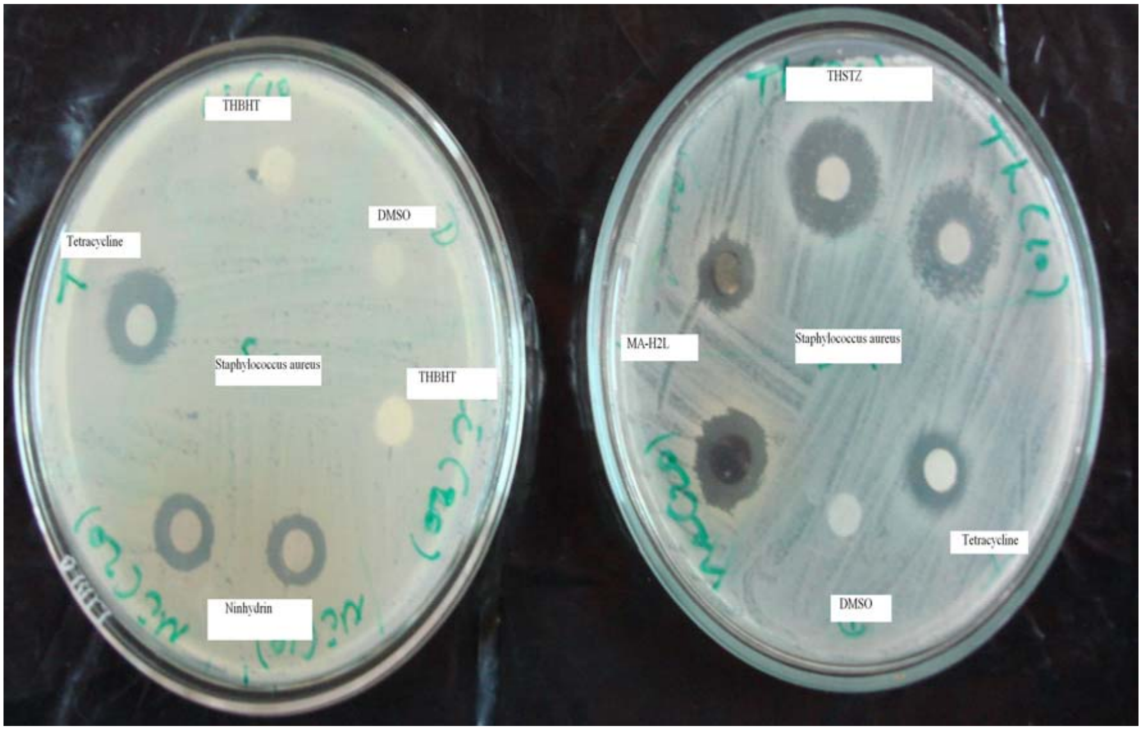
Appendix UV-Vis Spectrum of Ni(II) Complex in dil. Solution



Appendix Pictures showing inhibiting activities of the Compounds against the tested bacteria



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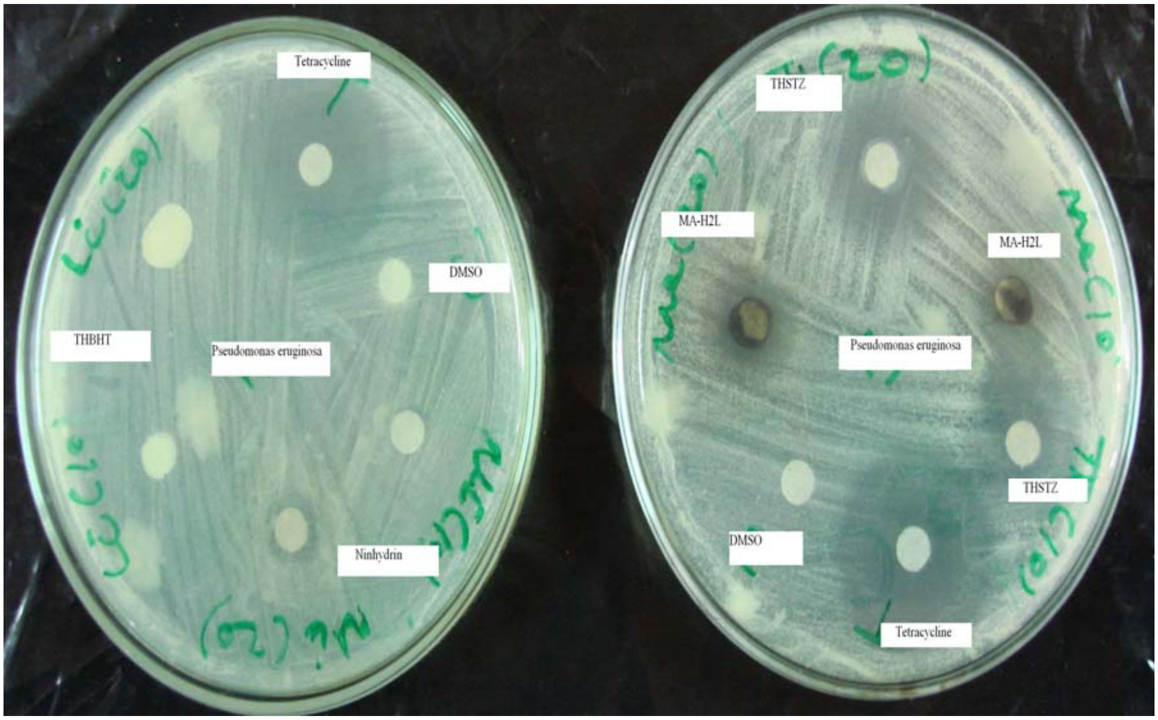
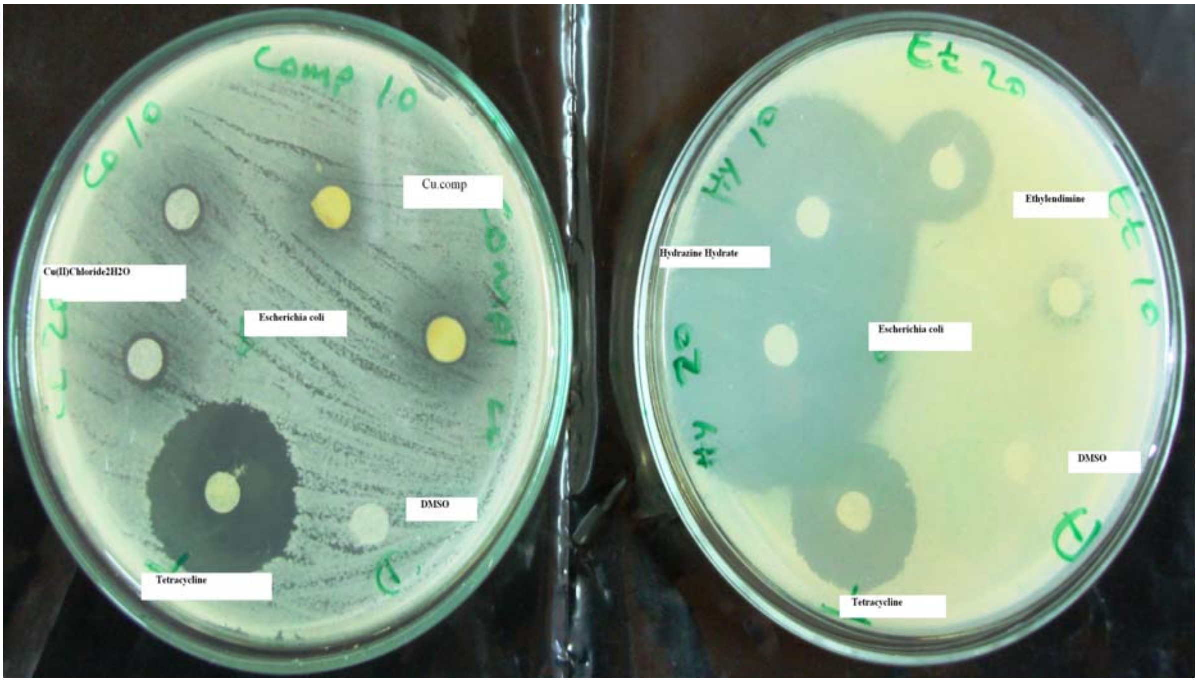
DMSO
Tetracycline
Staphylococcus aureus
THBHT
Ninhydrin
THSTZ
MA-H2L
Staphylococcus aureus
Tetracycline
DMSO


Tetracycline
DMSO
THBHT
Pseudomonas eruginosa
Ninhydrin
THSTZ
MA-H2L
MA-H2L
Pseudomonas eruginosa
THSTZ
DMSO
Tetracycline


Comp 1.0
Co 1.0
Cu.comp
Cu(II)Chloride2H2O
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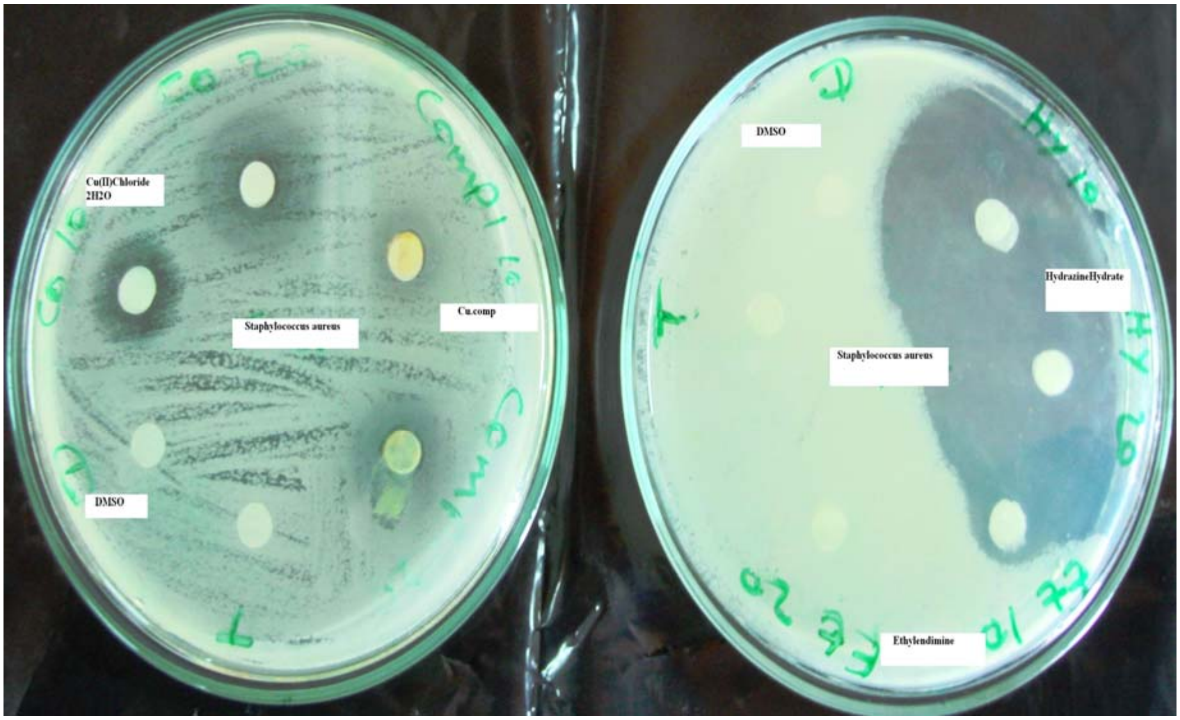
DMSO
Tetracycline
Et 20
Hy 10
Hydrazine Hydrate
Ethylendimine
Et 10
Escherichia coli
Hy 20
DMSO
Tetracycline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Comp 1.0
Cu(II)Chloride
2H2O
Co 1.0
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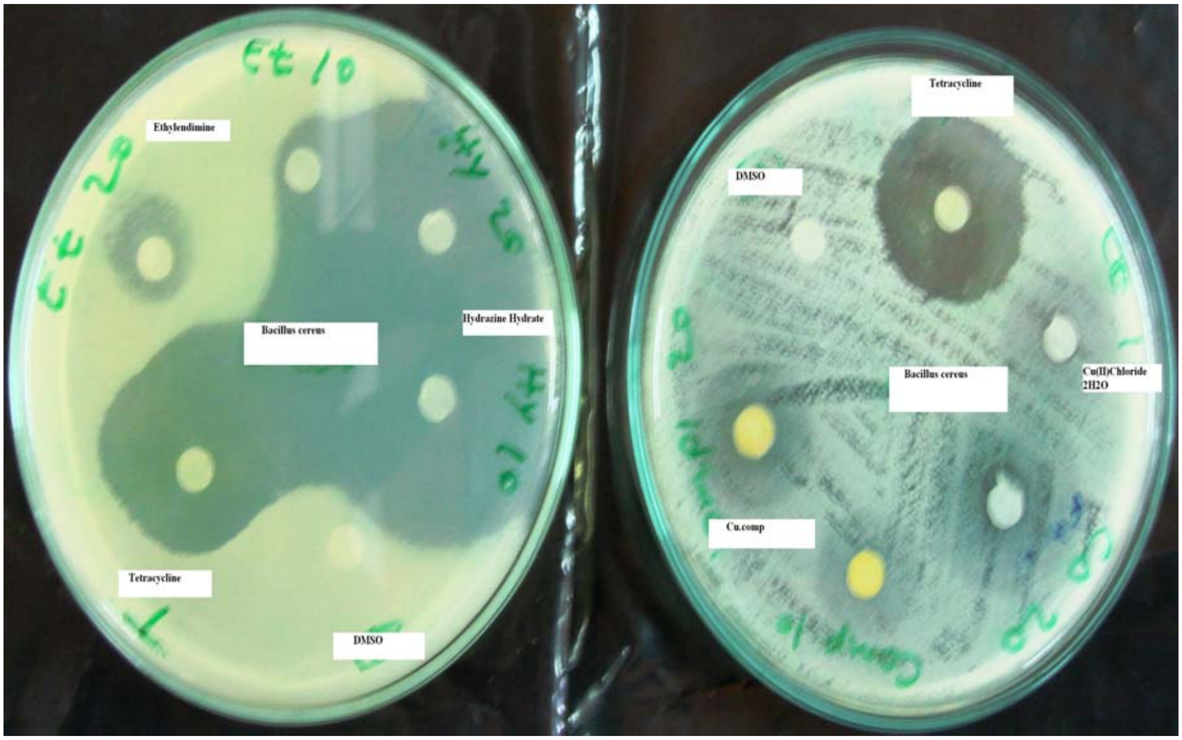
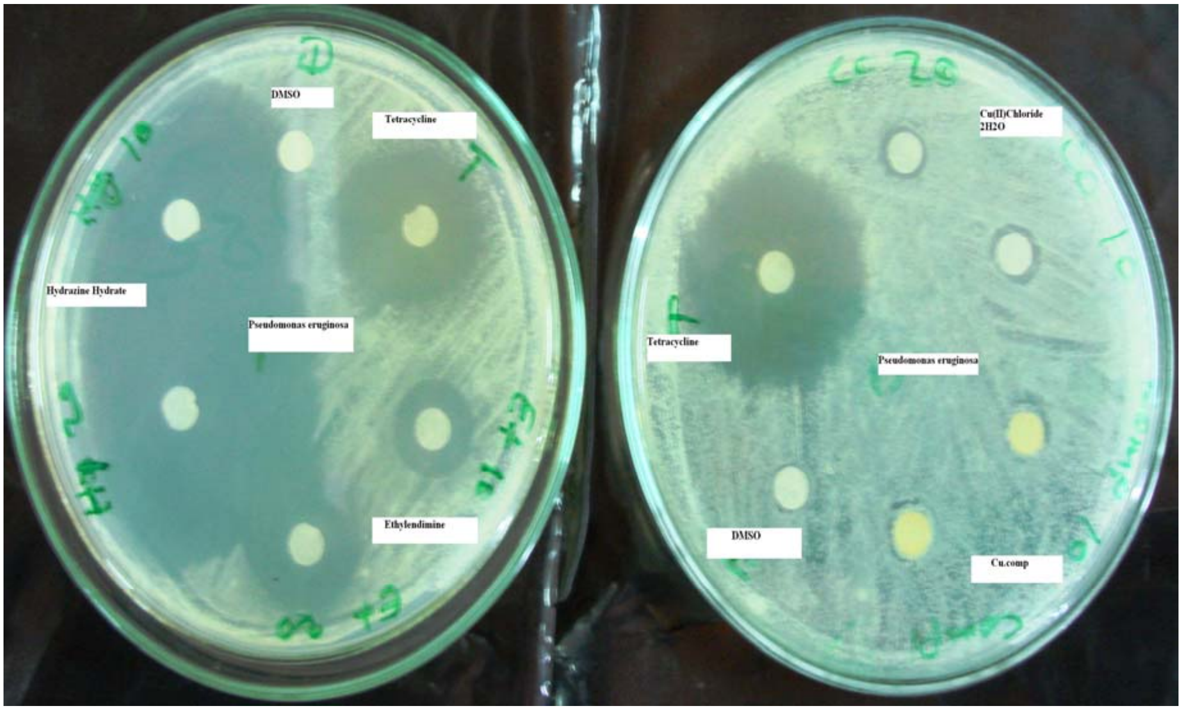
Cu.comp
DMSO
DMSO
Hy 10
HydrazineHydrate
Staphylococcus aureus
Hy 20
Et 10
Et 20
Ethylendimine


Ethylendimine
Hydrazine Hydrate
Bacillus cereus
Tetracycline
DMSO
Tetracycline
DMSO
Cu(II)Chloride 2H2O
Bacillus cereus
Cu.comp


DMSO
Tetracycline
Hydrazine Hydrate
Pseudomonas eruginosa
Ethylendimine
Cu(II)Chloride 2H2O
Tetracycline
Pseudomonas eruginosa
DMSO
Cu.comp