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Structural Elucidation of Cobalt (II) Complexes of 2-Imino-3-(2-hydroxyphenyl)-1-oxozolodin-4-one and Study of its Antimicrobial Relevance

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Abstract

Complex of Co(II) with heterocyclic ligand [2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one] has been synthesized and characterized with the help of elemental analysis, magnetic, ^{1}H-NMR, ^{13}C-NMR, IR and electronic spectral data. IR spectra exhibit the coordination of the ligands to the metal ion through deprotonated phenolic oxygen and heterocyclic nitrogen. All these studies reveal square planar geometry of Co (II) complexes. Synthesized compounds have also been screened against some micro-organisms *viz. Escherichia coli, S. aureus, Aspergillus niger* and *R. bataticola* by 'paper disc' techn ique. It has been observed that the antimicrobial activities of metal complexes are higher than that of the free ligands.

Keywords: Heterocyclic ligands; Oxazolodinone; Cobalt complexes; Antimicrobial studies; Electronic spectra; Co (II) complexes

Introduction

Metals have been used in the treatment of diseases of humans since ancient times. The Chinese were using elemental gold for the treatment of diseases, a practice known as chrysotherapy, as far back as 2500 B.C [1-5]. In more recent times, a stable metal coordination complex based on the element platinum, *cis*-[$PtCl_2(NH_3)_2$] (cisplatin), has become the most well-known of all metal based drugs and hundreds of articles have been published on the synthesis and activity of complexes derived from the parent cisplatin molecule. Since Rosenberg's initial discovery of cis-platin in 1969 many more examples of metal-containing drugs have been reported in the literature. Gold containing complexes such as auranofin are commonly used to treat rheumatoid arthritis. Radiopharmaceuticals based on metals such as technetium and rhenium is used in imaging and radiotherapy and ruthenium complexes have had some success as anticancer drugs.

Complexes containing gadolinium, cobalt, lithium, bismuth, iron, calcium, lanthanum, gallium, tin, arsenic, rhodium, copper, zinc, aluminium and lutetium have all been used in medicine [3].

Cobalt metal and its complexes

The word 'cobalt' is derived from the German 'Kobalt', from kobold, meaning 'goblin', a word used by miners for the ore of cobalt [2]. Cobalt occurs naturally as only one stable isotope, ^{59}Co. ^{60}Co is a commercially important radioisotope, used as a radioactive tracer and in the production of gamma rays. ^{60}Co is the isotope of primary importance to waste disposal and site remediation. ^{60}Co is an activation product of stable ^{59}Co, which is a common impurity in zircaloy and aluminium fuel cladding and in uranium metal fuel. ^{60}Co is a cobalt radioisotope most often used in medical tests, as a radiolabel for vitamin B_{12} uptake, and for the Schilling test. The Schilling test is a medical investigation used for patients with vitamin B_{12} deficiency. The purpose of the test is to determine whether the patient has pernicious anemia (Table 1).

Being transition element cobalt shows several variable valencies. In all oxidation states it forms stable complexes with diverse ligands of different stereo chemistries as shown below.

Transition metal complexes containing macro cycles are of considerable interest in terms of structural and coordination chemistry [6-22]. The chemical properties and structures of macro cyclic ligands can force metal ions to adopt unusual coordination numbers and geometries. Transition metal macro cyclic complexes have received much attention as an active part of metalloenzymes and as biomimetic model compounds due to their resemblance to natural proteins such as hemerythrin and enzymes. Aza-type ligands appear very promising for potential use as antifertile, antibacterial and antifungal agents as well as other biological properties. Transition metal complexes have received much attention as catalysts in oxidation and epoxidation processes. Structural factors such as ligand rigidity, the type of donor atoms and their disposition have been shown to play significant roles in determining the binding features of macro cyclic ligands toward metal ions [4]. Cobalt is the active centre of coenzymes called cobalamins, the vitamin B-12 is one of the most important example of the Co (II) complexes of macro cyclic ligand having corrin structure. As such it is an essential trace dietary mineral for all animals. Cobalt in inorganic form is also an active nutrient for bacteria, algae and fungi.

Hetrocyclic ligands with donors (N, O, S, etc.,) have structure similarities with natural biological systems and due to presence of imine group utilized in elucidating the mechanism of transformation of rasemination reaction in biological system. This may be attributed to their stability, biological activity and potential applications in many fields such as oxidation catalysis, electrochemistry, etc., Various studies which have shown a relationship between the metal ions and their metal complexes as antitumor and antimicrobial agents are a subject of great interest. The complexes of transition metals with heterocyclic ligands have been found to play a vital role in medicine, biological systems and industries. The complexes of Schiff's base formed by the condensation of 1-formyl-2-hydry-3-naphtholicarylamide with o-hydroxyl or o-methoxy aniline with Co (II), Ni (II), Cu (II) and Zn (II) are useful

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Oxidation state	Coordination number	Geometry of complexes
Co^{II}, d^7	3	Trigonal
	4	Tetrahedral
	4	Square planar
	5	Trigonal bipyramidal
	5	Square pyramidal
	6	Octahedral
	8	Dodecahedral
Co^{III}, d^6	4	Tetrahedral
	4	Square planar
	5	Square pyramidal
	5	Trigonal bipyramidal
	6	Octahedral

Table 1: Oxidation states and stereochemistry of cobalt.

as figments. Oxovanadium complexes have been found strongly active against some type of Leukaemia. Transition metal complexes derived from a number of amino acids have been reported to have biological activities [1].

Oxazolidinone selectively inhibits bacterial protein synthesis through binding to sites on the bacterial ribosome and prevents the formation of a functional 70S-initiation complex. Specifically, linezolid binds to a site on the bacterial 23S ribosomal RNA of the 50S sub-unit and prevents the formation of a functional 70S initiation complex which is an essential component of the bacterial translation process. Oxazolodinones are new class of totally synthetic antimicrobial agents against multidrug-resistant gram-positive and gram-negative bacteria, including methcill inresistant staphylococcus aureus (MRSA), staphylococcus epidermidis (MRSE), penicillin resist -ant streptococcus pneuonomiae (PRSP) and vancomycin-rsistant entrococci (VRE). The unique mechanism of action of oxazolidinones as attracted interest to develop derivatives with potent activity and broad spectrum [19].

Oxazolidinones have: A novel mechanism of action, selectively binding, uniquely binding to the 50S ribosomal subunit, inhibiting bacterial translation at the initiation phase of protein synthesis. Consequently, the drug would not show cross resistance with existing antibacterial agents.

Oxazolidinones, developed in recent past, with oxygen and nitrogen in penta-cyclic ring, have also been found to display profound wide spectrum of biological properties [21].

A survey of literature reveals plenty of reports on variety of ligands used for the synthesize, characterization and evaluation diverse biological properties of the metal complexes. On the transition metal complexes of oxazolidinones very little attempts have done. In view of scarce knowledge in the chemistry and antimicrobial potential of transition metal complexes with mentioned hetrocyclic organic ligands in general and non-availability of any reference on chemical and biological properties of Co (II) complexes with imino-oxazolidinones in particular which have two nitrogen's of imino and heterocyclic ring and oxygen donors, present research project which includes synthesis, characterization and antimicrobial study on Co(II) complex with 2-imino-3-(2-hydroxyphenyl)-1-oxazolidin-4-one ligand have been planned and proposed for investigations (Figure 1).

To synthesize, characterize and anti-microbial assay of cobalt (II) complex with 2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one,

To synthesize 2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one its C o (II) complex,

To characterize the new ligand and its Co (II) complex by elemental analysis, magnetic, spectral (IR, UV/Vis, ^{1}H NMR, ^{13}C NMR) analytical methods,

To study antimicrobial (bactericidal and fungicidal) activities of 2-imino-3-(2-hydroxyphenyl)-1-oxazolidin-4-one and its Co (II) complex against some Gram-positive and Gram-negative bacteria and fungi by using paper disc method.

Materials and Methods

The chemicals including chloroacetyl chloride, o-aminophenol, KCNO, $CoCl_2$ and $Co(NO_3)_2$ used in the present synthetic work were libratory reagents (BDH, E merk, Coligien). All solvents such as benzene, acetone, ether, etc used in TLC studies were HPLC grade.

Instruments and apparatus

The ^{1}H and ^{13}C NMR spectrum of the ligand was recorded in DMSO on Bruker Ultra shield TM 400 spectrometer using TMS as internal standard at Addis Ababa University. The infrared spectra were recorded on Fourier Transform Infrared (FT-IR) spectrophotometer (Prestige- 21) in the range 4000 cm^{-1}-400 cm^{-1} in KBr medium; C, H, N and S elemental analyses were done at IIT, Roorkee (India). The melting points of the synthesized compounds were determined in an open glass capillaries using Bibby Sterilin LTD, ST150SA, UK melting point apparatus. Electronic spectral measurements were done using UV/Vis-SP65 SYANO spectrophotometer in 200-900 nm range. Magnetic susceptibility measurements were done on MSB-AUTO, (Sherwood Scientific) magnetic balance at Addis Ababa University. The molar conductivity measurement was carried out using Jenway digital conductivity meter (UK) at Haramaya University. ^{1}H-NMR and ^{13}C-NMR, FT-IR, magnetic susceptibility measurements and elemental analysis were done outside Haramaya University whereas; other instrumental analysis and synthetic works were done in the Chemistry Department research laboratories of Haramaya University. Antimicrobial assay work of the synthesized compounds was carried out at the Plant science Department, Haramaya University Experimental Procedures.

Synthesis of 2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one

Equimolar solutions of the reactants 2-hydroxy aryl chloroacetanilide (3 g) and potassium cyanate (1.75 g) were mixed in methanol together in 1:1 ratio and the reaction mixture was refluxed for about 7 hrs and filtered to remove KCl. Solvent of the filtrate was evaporated on water bath and residue was washed with water repeatedly to ensure complete removal of KCl, dried and crystallized from ethanol. Crystals of product were dried in air.

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Preparation of cobalt (II) complexes

All the cobalt (II) complexes were prepared by a common procedure. Mixture containing cobalt each of cobalt (II) salts ($CoCl_2/Co(NO_3)_2$) and ligand (iminoxazolidinone) in 2:3 molar ratio in acetone were refluxed for 2-3 h. The reaction mixtures were concentrated on water bath and allowed to cool at room temperature (Scheme 1). Precipitates products were filtered out and washed with water and ethanol (small amount) successively and dried in air.

Test of purity of the products

The purity of the ligand and its metal complexes were tested by using thin-layer chromatography (TLC) on silica gel thin layers using and two component solvent systems (AcOEt-n-hexane, 9:2 2:1, v/v). The entire product showed single spot on thin-layers in all the one component and (AcOEt-hexane (9:2, v/v). The complex sample showed two spots in (AcOET-hexane 2:1, v/v). This sample was washed with AcOEt-hexane (2:1, v'/v) solvents repeatedly to obtain pure product.

Chloride analysis

0.05 gm of complexes were decomposed with conc. HNO_3 and diluted. To the resultant solution of the complexes aqueous solution of $AgNO_3$ was added. Again to the solution of each complex in methanol on $AgNO_3$ solution was added. In both chloride and conductance treatments there was no precipitate of AgCl. This indicates absence of chloride and nitrate inside and outside (on outer sphere) of the coordination zone of the metal. This result shows that complexes from both salts are similar.

Determination of elemental composition (Determination of metal content in the complexes)

The metal content in the complex was determined spectroscopically using Atomic Absorption Spectroscopy (AAS). The experimental percentage of the metal in the complexes was obtained from the AAS data using the formula:

$$M(\%) = Concentration\,(\mathrm{PPm}) \times \frac{100}{1000}$$

40 mg each of Co (II) complexes were placed in two different clean and dry beakers to each of which 20 ml of conc. HNO_3 was added and the contents were heated gently in a hood until a few drops remained in each beaker. Then 10 ml of additional conc. HNO_3 was added to each beaker and heated slowly until a few drops remained. This procedure was repeated until all beakers hold same colored solution then the remaining small amount at the bottom of the beaker was diluted with distilled water in 100 ml flask. These solutions were subjected to AAS studies. Based on the absorbance data the concentration of Co (II) in the complexes was calculated.

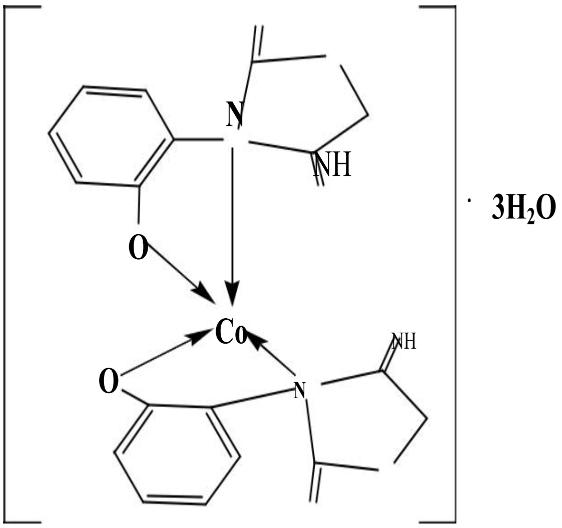


Figure 1: Structure of 2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one cobalt.

Elemental analyses of C, H and N

Elemental analyses for C, H and N contents of all the synthesized compounds was conducted spectrophotometer (Prestige- 21) at chemistry departments of IIT, Roorkee, (India).

Physico-chemical methods of characterization coductrometric measurement

Conductometric measurements on the complexes were done by dissolving 50 mg of each complex in 10 ml organic solvent (methanol DMSO) and the molar conductivities of their solutions were measured at room temperature by carefully adjusting instrument by 0.1 mol of KCl solution.

Magnetic susceptibility measurements

The magnetic susceptibility measurements on the complexes were done to know their magnetic nature and stereochemistry at room temperature on a simple Gouy type balance at 22°C. The effective magnetic moment was calculated using the equations:

$$X_M = X\ (\mathrm{Mol.Wt}) \qquad \text{(i)}$$

$$X'_m = X_M\text{-Diamagnetic correction} \qquad \text{(ii)}$$

$$\mu_{eff} = 2.84\ (X'_m T)^{1/2}\ \mathrm{BM} \qquad \text{(iii)}$$

Where, X=Gram susceptibility, T=absolute temperature, X_M=Molar susceptibility, X'_m=Effective molar susceptibility.

Spectral measurement

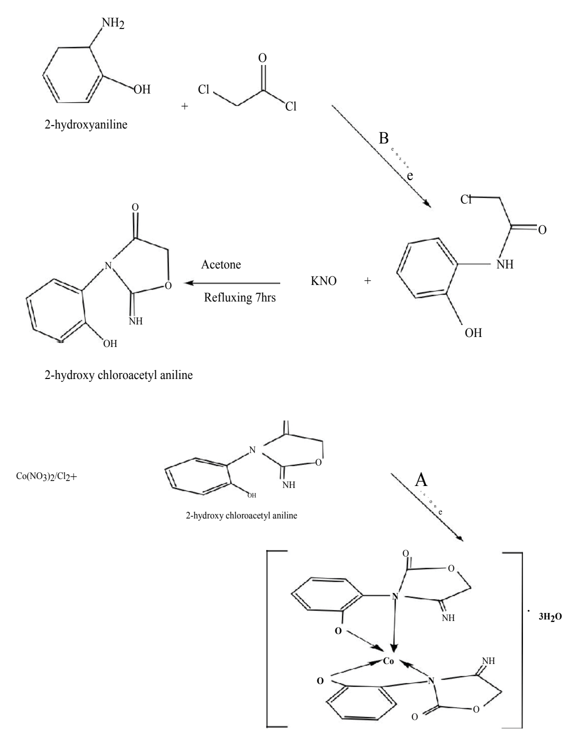
Uv-visible absorption spectra on the solutions of the complexes in 200-900 nm regions were recorded in methanol and DMSO to identify and determine energies of d-d electronic transitions and electronic charge transfers.

FT-IR spectra of the ligand and its complexes recorded in 400 cm^{-1}-200 cm^{-1} range in KBr medium on FTIR-Shimadzy Spectrometer were used to identify the presence of functional groups and to receive valuable information regarding the coordinating sites of the ligand in the metal complexes [23,24].

For ^1H-NMR, ^{13}C-NMR and DEPT-135 spectra 10 mg of each complex in DMSO solvent were recorded on Brucker 400 MHz Advance spectrometer and the chemical shift value are expressed in (δ, ppm) and reported in parts per million down field using internal reference Tetra methyl Silane (TMS).

Antimicrobial activity studies

The antimicrobial assay was done against two fungi (*E. coli* and *R. bataicola*) and two bacteria (*S. aureus* and *A. niger)* by paper disc diffusion method. Mueller Hinton agar (MHA) was used to cultivate *S. aureus* and *E. coli* whereas *A. niger* and *R. bataticola* were cultured on Potato Dextrose agar (PDA) medium. The degree of bactericidal and fungicidal activities were determined by measuring diameter of inhibition zone and compared with the reference standard antibiotic mixture and Beastie [9].



Scheme 1: Synthesis of 2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one.

Preparation of inoculums: The test bacterial strains' were transferred from the stock cultures as streaked on Mueller Hinton agar (MHA) and incubated for 24 hrs at 37°C. Well separated bacteria colonies were then used as inoculums. Incubating loop used to autoclaved MHA that was cooled to about 37°C in water bath and mixed by gently swirling the flasks. The medium was poured to sterile petri plates allowed to solidify and used for the bio test**.** A fresh culture of inoculums of each culture was streaked on Mueller Hinton agar (MHA) in a petridish. 10 µl and 20 µl solutions containing 6 mg/ml synthesized ligand and its Co (II) complexes were impregnated using micropipette on paper disc of 6 mm in diameter. Both samples were applied in triplicate.

Preparation of test solution: Three samples were prepared for fungicidal and bacterial test labelled as 9, 11 and 12 respectively. The synthesized ligands and their Co (II) complexes solutions were prepared at con centration of 6 mg/ml by dissolving them in DMSO (dimethylsulfoxide). Zones of inhibition were measured after 36 h of incubation. Finally activity of all synthesized ligand and its complexes on fungus and bacteria's were determined by measuring inhibition zone and taking average of measured values.

Results and Discussion

Physical characteristics of the ligand and its complex

Elemental, metal contents and conductance data are presented in Tables 2-4 respectively. Elemental contents including carbon, hydrogen, nitrogen and metal and molar conductance values of the complexes are in fair agreement with the proposed chemical composition of the products. Metal concentration in the complexes calculated by using standard calibration curve (Appendices 1-4). From the Table 2 it is clear that the contents of each element C, H and N contents in the ligand greater than their content in complexes this indicate the formation of bond between the ligand and cobalt (II).

Physico-chemical characterization (Colour, formula and melting point analyses)

In Table 3 change of colour of ligand (2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one) and rise of melting point when mixed with metal salts shows the formation complexes. These complexes are colourful due to existence of spectra in UV/Vis region as showed in Appendices 5 and 6.

Determination of metal content by AAS

After complete digestion of complexes with nitric acid all the organic parts removed and both the two beakers containing yellow colored solutions showed solution of Co (II) salt. These solutions showed absorbance in range from 0.060 and 0.065. The content of Co (II) in each complex was in fair agreement with the proposed chemical composition of the products. The metal contents were in the range 11.59 and 11.92 showed the existence of Co (II) in synthesized complexes.

Conductometric study

From Table 5 the small molar conductance value of all complexes falling in the range 4.42 and 4.50 $\Omega^{-1}cm^2mol^{-1}$ indicated that all complexes are non-electrolytic in nature. This is due to absence of both anions (chloride and nitrate) in outer coordination sphere.

Magnetic studies

Table 6 shows that Magnetic moments of the complexes lying in 1.45-1.48 BM range are neither corresponding to spin-free S=3/2 or S=1/2 configurations of d^7 Co (II). From these values of magnetic moments it is well clear that ligand is strong field ligand which forced maximum pairing of d-shell electrons of the metal. The observed magnetic moment values close to 1.73 BM of S=1/2 complexes involving strong field ligand however indicate that all the four complexes have only one spin-free electron in square planar geometries whereas lower magnetic moments than spin-free value of 1.73 BM could most probably be due their ligand bridged dimeric structures involving metal-metal interactions.

Electronic spectra studies of complexes

As shown in Appendices 5 and 6, electronic spectra of complexes displayed one, two or three d-d transition bands of low energy in 14706-27027 cm^{-1} region and two bands of high energy in 31250-43478 cm^{-1} range as summarized in Table 7 below. The low energy bands attributed to $^2A_{1g}$, $^2B_{2g}$ and ^2E_g transitions from $^2B_{1g}$ ground state of square planar geometry as revealed by magnetic data; broad peak structure of $^2B_{1g} \rightarrow {}^2B_{2g}$ electronic transition band peak could be due to its mixing either with closely spaced peak of $^2B_{1g} \rightarrow {}^2A_{1g}$ or $^2B_{1g} \rightarrow {}^2E_g$ transition. The high energy peaks are assigned to metal to ligand or vice versa charge transfers.

IR spectra studies

As stated in Table 8 interpreted from electronic spectra from Appendices 2-4, the perusal of infrared spectra of 2-imino-3-(2-hydroxyphenyl)-1-oxazolidin-4-one and its complexes reveals considerable lowering of ν C-N-C band of the ligand (1372 cm^{-1}) in the complexes (1279-1282 cm^{-1}) disappearance of νOH peak of ligand (3448 cm^{-1}) in the complexes and appearance of a new band at 533 cm^{-1} and 538 cm^{-1} in the complexes are the clear evidences of the coordination of deprotonated oxygen of phenolic group and appearance of a new band at 446 cm^{-1} and 423 cm^{-1} in the complex spectra. These spectral observations lead us to propose the coordination of heterocyclic nitrogen with cobalt (II). The Lattice water displayed symmetric and

Compounds	Elemental Analysis Calculated (Found) %				
	C	H	N	S	M%
$C_9H_7N_2O_3(L_2)$		4.19-4.2	11.72-12.24	-	-
$[CoL_2].3H_2O$		4.04-4.02	11.31-10.88	-	11.59-12.88
$[CoL'_2].3H_2O$	43.64	4.04-4.47	11.31-10.83	-	11.92-11.6

Where, L=C9H7N2O3

Table 2: Elemental analysis data of ligands and their complexes.

Compounds	M. Wt	Colour	M. Pt (°C)
$C_7H_9NO_3$ (L_2)	191	Bronze	90°C
$[CoL_2].3H_2O$	495	Brown	29
$[CoL_2\ ].3H_2O$	495	Blue black	126

$L=C_9H_7N_2O_3$ (L_2)

Table 3: Colour, Formula and Melting point.

Compound	Mol.wt	Conc. (ppm)	Absorbance	Calculated	Found
$[CoL'_2].3H_2O$	495	16.23	0.065	11.59	12.88
$[CoL'_2].3H_2O$	450.7	11.13	0.06	11.92	11.6

Table 4: AAS data for the M-L complexes.

Compounds	Conductance of Complex(μS)	Specific conductance $\times 10^6$ (ohm^{-1} cm^2 mol^{-1})	Molar conductance (Ω^{-11} cm^2 mol^{-1})	Electrolytic nature
$[CoL'_2].3H_2O$	2.22	2.19	4.42	Non electrolyte
$[CoL_2].3H_2O$	2.25	2.23	4.5	Non electrolyte

Cell constant (k)=0.99, Conductance of DMSO and methanol=3×10^{-8} and $4.4 \times 10^{-7}\ \Omega^{-1}$

Table 5: Molar Conductance data.

Compounds	M Wt	Gram susceptibility $\times 10^6$ (cgs)	Molar susceptibility $\times 10^6$ (XM) (cgs)	Diamagnetic correction $\times 10^6$ (cgs)	Effective susceptibility × Mcorr $\times 10^6$	Eff. Magnetic moment (μ eff) (BM)
$[CoL_2].3H_2O$	495	1.457	721.22	-166.28	887.5	1.45
$[CoL_2].3H_2O$	495	1.527	755.87	-166.28	922.15	1.48

Table 6: Magnetic Susceptibility and magnetic moment measurement data.

Complexes	Band frequency (cm^{-1})	Assignment	
$[CoL_2].H_2O$	23255	$^2B_{1g}{\to}^2B_{2g}$	
		Charge transfer	
	33893	Ligand	Metal
	43478		
$[CoL_2].3H_2O$	20833	$^2B_{1g}{\to}^2B_{2g}$	
	24390	$^2B_{1g}{\to}^2E_g$	
	34482	Charge transfer	
	43472	Ligand→Metal	

Table 7: Electronic spectra bands frequencies (cm^{-1}) with their assignment.

anti-symmetric stretching and bending vibrations at 3380 and 1645-1700 cm^{-1} region respectively. Doublet peak structure shows mixing of two closely spaced peaks from the infrared studies. It is infrared that the ligands is bidentate and coordinate with the metal through their deprotonated phenolic oxygen and heterocyclic nitrogen. In all the four complexes Co(II) salts anions (chloride and nitrate) do not play any role in coordination.

^{1}H-NMR and ^{13}C-NMR studies

Generally structural elucidation of ligand was determined by using ^{1}H NMR, ^{13}C NMR and DEPT spectra shown on Appendices 7 and 8. As shown in Table 9 NMR spectra of oxazolodinone displayed signals in δ 3.85 - δ 4.30, δ 8.10 - δ 9.85, δ 8.10- δ 8.75 and δ 5.55 - δ 6.20 regions due to their OCH_3, OH (phenolic), CH-N (ring) and $-S-CH_2$ group respectively; benzene protons exhibit multiple signals in δ 6.10 -7.60 ppm region and its ^{13}C-NMR characteristic signals observed in δ 56 - 66, δ 139-152, δ 55-56, δ 166-191 and δ 30-34 respectively whereas aromatic signals are observed in δ 109 - 158 region.

Structural elucidation

Based on the combined analysis spectral, magnetic and conductance results the structure of all complexes in square planar geometry are proposed as shown below.

Antimicrobial activities

Antimicrobial assay data in Table 10 exhibit dose dependent toxicity against both bacteria and *A. niger* fungus. Both ligand and Co (II) complex are inactive against *R. bataticola* fungus. $CoCl_2$ oxazolidinone complex however, is inactive against *S. aureuos* and bacterium and *A. niger* respectively. The complex showed best in toxic effect against *E. coli* in comparison of other microbial.

^{v}*C-O-C* (ring)	^{v}C-S-C (ring)	^{v} N-H secondary ammine	^{v} C=N	^{v}C-N-C $_{ring}$	Phenolic v OH	Benzene v C=C	^{v} C-H	Ortho disubtitution	^{v}M-O	^{v}M-N	*Lattice water*
1106	-	3341	1554	1372	3448	1455	2983	749	-	-	-
						1600					
1108	-	3367	1552	1279	-	1458	2982	749	533	446d	3448
						1595					1654
											1700
1103	-	3343	1550	1282	-	1452	-	748	538	422	3449
			Br								1645
											1699

Where, L=$C_9H_7N_2O_3$, br=broad and d=doublet

Table 8: Principal IR frequencies with their assignments for ligands and their complexes.

Compounds	Chemical shift (δ, ppm)	^{1}H NMR Assignment	^{13}C NMR	
			Chemical shift (δ, ppm)	**Assignment**
$C_9H_7N_2O_3$(L)	6.26-6.60	Benzene ring H	115-123	Benzene carbon (C-H)
	5.0	Phenoloic OH	129	Benzene carbon (C-N)
	5.14	Methylene with 1-α -N- and 1-α C-O	143.5	Benzene carbon (C-O)
	3.4	Methylene pyrrollidine	165	Pyrrollidine (C=NH)
	4.4	Pyrrollidine N-H		

Table 9: ^{1}H NMR and ^{13}C NMR spectral data of the ligand.

Metrics	Dose (µL)	Inhabition zone (mm)			
		Fungi		**Bacteria**	
		E. coli	***R. bataticola***	***S. aureus***	***A. niger***
$C_9H_7N_2O_3$ (L')	10	14	-	-	9
	20	17.5	-	-	11
$[CoL'_2].3H_2O$	10	12	-	9	8
	20	13	-	10	8
Ref. 1	10	23	-	-	22
	20	27	-	-	26.5
Ref. 2	10	-	-	18	-
	20		-	20	-

Where, Ref. 1=Mixture of Chloramphenicol and streptomycin Ref. 2=Bavastin

Table 10: Inhibition zone of bacterial and fungal growth in (mm).

Summary and Conclusion

These heterocyclic ligands 2-imino-3-(2-hydroxyphenyl)-1-oxazolodin-4-one was synthesized by the cyclization of 2-hydroxy chloroacetyl aniline obtained by reaction o-aminophenol and chloroacetylchloride, with KCNO respectively. This ligand was fully characterized by elemental analysis, melting point, IR, ^{1}HNMR and ^{1}CNMR. This heterocyclic ligand was successfully coordinated to cobalt (II) ions to form its corresponding complex. The comparison of the IR spectra of the synthesized ligand and its Co (II) complex indicated bi-ligancy of the ligand through its deprotonated phenolic oxygen and heterocyclic nitrogen as exhibited by disappearance and lowering in frequencies of phenolic (OH) and heterocyclic C-N-C groups of ligand also gave additional evidence on the formation of this complex. Phenolic (OH) and heterocyclic C-N- group of ligand in its complex by appearance of new bands due to *ν*(M-N) and *ν*(M-O) in the spectra of metal complex. The complex was formulated as mononuclear species on the basis of chemical analysis, molar conductivity measurements, electronic; IR and NMR spectral data. The electronic spectra indicate a low spin square-planar geometry for both the cobalt (II) complexes. The effective magnetic moment of the complex, which lies in the range of 1.45- 1.48 B.M close to one unpaired electron for low spin square-planar cobalt (II) complexes. However lower magnetic than spin-only value 1.73 BM reveal ligand bridged dimeric structure of all the complex involving metal-metal interaction. Antimicrobial assay data of the ligand and complex revealed moderate toxicity of almost all the synthesized compounds against both bacteria and *A. niger* fungus but all products are inactive against *R. bataicola* fungus.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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