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Synthesis, Spectroscopic Characterization, Antimicrobial and Antioxidant Properties of Some Metal(II) Complexes of Mixed Ligands-Riboflavin and 4-Aminobenzoic Acid

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ABSTRACT

Mn(II), Fe(II), Co(II), Ni(II), Cu(II) and Zn(II) complexes of mixed ligands, Riboflavin (HL) and 4-aminobenzoic acid (HL¹) were synthesized and characterized by percentage metal, infrared and electronic (solid reflectance) spectroscopies, room temperature magnetic moments, melting points and conductance measurements. The conductance measurements in DMSO and percentage metal analysis indicated that all the metal(II) complexes were covalent and analyzed as [M(HL)(HL¹)X].aH₂O, where X = Cl₂/SO₄. Infrared spectra data confirmed that coordination was via the oxygen atoms of two hydroxyl groups in Riboflavin, and the carboxylate oxygen atoms in 4-aminobenzoic acid respectively. Furthermore, electronic spectra data indicated that all the metal(II) complexes adopted octahedral geometry, while room temperature magnetic moment measurements indicated spin-crossover, that is, high spin ⇌ low spin equilibrium for all the complexes with the exceptions of the Cu(II) and Zn(II) complexes. *In-vitro* antimicrobial activities of the metal(II) complexes, riboflavin and p-aminobenzoic acid against *Escherichia spp*, *Proteus mirabilis*, *Streptococcus pyogenes*, *Candida albicans*, *Salmonella sp*, *Streptococcus sp*, *Bacillus spp*, *Staphylococcus sp* and *Pseudomonas spp* revealed that all the metal(II) complexes and ligands were active against *Pseudomonas sp* with inhibitory zones range of 7.0-11.0 mm. The antioxidant studies on the metal complexes showed that the Zn(II) complex had the best antioxidant activity of about 62 percentage inhibition, which was about twice the percentage inhibition of the standards, ascorbic acid and α-tocopherol.

INTRODUCTION

Recent research by our group and others are focused on drug metal complexes with the sole aim of discovering more effective chemotherapeutic agents to fight infectious diseases [1-8]. Furthermore, Riboflavin (Vitamin B₂), a water-soluble vitamin is converted to flavinadenine dinucleotide and flavin mononucleotide in microbes for production of energy [9-12]. Riboflavin is also used in treatment of neonatal jaundice and prevention of migraine and its deficiency results in sore throat, hyperemia, and edema of the pharyngeal and normocytic anemia [13]. In addition, Riboflavin is a multidentate ligand with imine nitrogen atoms, ketone oxygen atoms and hydroxy groups, which act as coordinating sites for various metal ions [10].

4-aminobenzoic acid is chosen as a secondary ligand due to its importance as a co-factor of the vitamin B complex and as a non-protein amino acid widely used by nature [14].

Thus, our aims are to synthesize, characterize and investigate the magnetic properties of the novel metal(II) complexes of mixed ligands, Riboflavin and 4-aminobenzoic acid for possible magnetic interactions such as spin-crossover, antiferro magnetism

and ferromagnetism. In addition, the suitability of these metal complexes as broad-spectrum antimicrobial agents *in-vitro* will be verified, as well as the inhibitory capacity of the metal complexes as antioxidants. This is a continuation of the research activities of our group on potentials of various metal(II) complexes as chemotherapeutic agents against infections and cancers [4-7].

EXPERIMENTAL

Material and reagents

Reagent grade Riboflavin, 4-aminobenzoic acid, 1,10-phenanthroline, Ascorbic acid, α -tocopherol, manganese(II) chloride tetrahydrate, Iron(II) tetraoxosulphate (VI) heptahydrate, Cobalt(II) chloride hexahydrate, Nickel(II) chloride hexahydrate, Copper(II) chloride dihydrate and Zinc(II) tetraoxosulphate (VI) heptahydrate were obtained from Aldrich and BDH chemicals and were used as received, while solvents were purified by distillation.

Physical measurement

The solid reflectance spectra of the metal complexes were recorded on a Perkin-Elmer λ 25 spectrophotometer and infrared spectra were recorded as KBr disc on a Perkin-Elmer FT-IR spectrum BX spectrometer in the range 4000-400 cm^{-1} . The room temperature magnetic susceptibilities at 303K were measured on Sherwood susceptibility Balance MSB Mark 1 and diamagnetic corrections were calculated using Pascal's constant. Melting points were determined with Mel-Temp electrothermal machine, while molar conductivity measurement of 1×10^{-3} M solutions in DMSO were obtained using electrochemical analyzer consort C933, and percentage metal was determined by complexometric titration using EDTA.

Antimicrobial assay

The assay was carried out on the ligands and their metal(II) complexes using the agar diffusion technique. The surface of the agar in a Petri dish was uniformly inoculated with 0.2 mL of 18 hours old test bacterial culture of *Escherichia coli* (Typed strain), *Proteus mirabilis*, *Streptococcus pyogenes*, *Candida albicans*, *Salmonella sp*, *Streptococcus sp* (Blood), *Bacillus sp* (food strain), *Staphylococcus sp*, *Pseudomonas sp* (Clinical isolate), *Pseudomonas sp* (environmental strain), *Bacillus sp* (environmental strain) and *Escherichia coli* (clinical strain). Using a sterile cork borer, 5 mm wells were bored into the agar. Then 0.06 mL, 10 mg/mL concentration of each metal complex in DMSO was introduced into the wells and the plates were allowed to stand on the bench for 30 minutes before incubation at 37°C for 24 hours after which inhibitory zones (in mm) were taken as a measure of antimicrobial activity. The experiments were conducted in duplicates and streptomycin sulphate was used as a reference drug.

Antioxidant assay

Ferrous ion-chelating ability: The ferrous ion-chelating ability was determined by the standard colorimetric method [15]. The reaction mixture contained 1 mL of 1, 10-phenanthroline (50 mg in 100 mL of methanol), 1 mL $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ (400 μM) and 1 mL of sample solution of the same concentration (1.0 mg/mL) and the reaction mixture was finally mixed with 2 mL of methanol. The mixture was then incubated at room temperature for 15 minutes, after which the absorbance of the sample was read at 510 nm spectrophotometrically. 2 mL of Methanol with 1 mL of 1,10-phenanthroline (50 mg in 100 mL of methanol) and 1 mL $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ (400 μM) solution was used as blank (control), while α -tocopherol and ascorbic acid were used as the standard. All tests were run in triplicate and percentage scavenging inhibition of ferrous ion-chelating ability was expressed as:

$$\% \text{ scavenging inhibition} = (A_c - A) / A_c \times 100 \quad (1)$$

Where, A_c is the Absorbance of blank (control) reaction, A is the Absorbance of sample solution or the Absorbance of standard respectively.

SYNTHESIS

Preparation of $[\text{Mn}(\text{HL})(\text{HL}^1)\text{Cl}_2] \cdot \text{H}_2\text{O}$

This complex was prepared by the addition of 0.42 g (2.12×10^{-3} moles) of $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ to a stirring solution of 0.80 g riboflavin (2.13×10^{-3} moles, HL) and 0.29 g of 4-aminobenzoic acid (2.11×10^{-3} moles, HL^1) in 50 mL of 70% methanol. The resulting homogenous solution was buffered with 4 drops of triethylamine from a pH of 4 to 9, and stirred for additional thirty minutes. The resulting cloudy solution was refluxed at 50°C for 6 hours during which the product formed. The yellow precipitate obtained was filtered, washed with 70% methanol and dried over silica gel. The same method was used for the preparation of the Fe(II), Zn(II), Co(II), Ni(II) and Cu(II) complexes from their sulphates and chloride salts respectively.

RESULTS AND DISCUSSION

The reaction of Riboflavin (HL) and 4-aminobenzoic acid (HL^1) with the metal(II) chlorides (Mn, Ni, Cu and Co) and metal(II) sulphates (Fe and Zn) respectively gave coloured complexes of moderate to good yields according to equation 2-3.



(When M= Fe, Zn; n = a = 7)



(When M =Mn, Cu, n = 4, a = 1, b = 3; Co, n = 6, a = 2, b = 4; M = Ni, n = 6, a = 1, b = 5)

The formation of the complexes was confirmed by % metal analysis which was in close agreement with the theoretical values. Furthermore, the ligands, Riboflavin(HL) and 4-aminobenzoic acid(HL¹) decomposed at 280°C and melted at 187-189°C respectively, whereas all the complexes decomposed in the range 200-254°C confirming coordination. Attempts to isolate single crystal for X-ray measurements were not successful. However, % metal analysis, IR and electronic spectroscopies, conductance and room temperature magnetic moments were used to obtain the formula mass of the complexes and their plausible geometry (Table 1).

Solubility and molar conductance measurements

The complexes exhibited good solubility in DMSO but were slightly soluble in ethanol, methanol, nitromethane and chloroform. Thus, their molar conductivities were determined in DMSO with values in the range 9.33 – 19.69 Ω⁻¹ cm² mol⁻¹ confirming their covalent nature, since values in the range 60-118 ohm⁻¹ cm² mol⁻¹ is expected for 1:1 electrolyte^[16].

Table 1: Analytical data of the ligands and their metal(II) complexes.

Complexes	Formula Mass	Color	M.pt(°C)	%Yield	%M(Exp)	Λ _M	
HL	376.36	Orange	280*	-	-	-	
HL ¹	137.14	Cream	187-189	-	-	-	D
[Mn(HL)(HL ¹)Cl ₂].H ₂ O	675.44	Yellow	248*	50	8.13(8.80)	12.33	3.74
[Fe(HL)(HL ¹)SO ₄]	663.35	Yellow	250*	78	8.42(8.31)	12.17	3.30
[Co(HL)(HL ¹)Cl ₂].2H ₂ O	679.43	Yellow	252*	57	8.67(8.77)	9.33	4.09
[Ni(HL)(HL ¹)Cl ₂].H ₂ O	667.87	Yellow	253*	54	8.88(8.74)	10.73	2.58
[Cu(HL)(HL ¹)Cl ₂].H ₂ O	662.85	Jasmine	200*	60	9.79(9.72)	19.69	2.27
Zn(HL)(HL ¹)SO ₄]	674.87	Yellow	254*	73	9.69(9.73)	11.00	0.80

HL = Riboflavin, HL¹ = 4-aminobenzoic acid, * = decomposition temperature, Exp = experimental, Λ_M = Molar conductance (ohm⁻¹cm²mol⁻¹); μ_{eff} = Effective magnetic moments, M.pt. = Melting point, D = diamagnetic

Electronic spectra and magnetic moments

The ultraviolet spectra of the compounds were characterized by strong absorption maxima 27.31-29.76 kK, 34.84 kK and 48.31 kK assigned to n→π*, π→π* and charge transfer respectively (Table 2). The Mn(II) complex had two absorption bands at 11.67 kK and 22.37 kK assigned to ⁶A_{1g}→⁴E_g and ²T_{2g}→²T_{1g} transitions respectively, consistent with high spin and low spin octahedral geometry. The effective magnetic moment of high spin Mn(II) complexes are expected to be close to the spin-only value at 5.90 B. M since the ground term is ⁶A_{1g} and thus, there is no orbital contribution, whereas low spin octahedral Mn(II) have moments of about 2.0 B.M. Consequently, an observed moment of 3.74 B.M. was indicative of spin equilibrium between the high spin and low spin octahedral geometry^[7,17]. The Fe(II) complex had two absorption bands at 18.73 kK and 24.39 kK typical of 6-coordinate, high spin and low spin octahedral geometry and were assigned to ⁵T_{2g}→⁵E_g and ¹A_{1g}→¹T_{2g} transitions. Moments in the range 5.0-5.6 B.M were reported for high spin octahedral Fe(II) while low spin octahedral Fe(II) were diamagnetic. In this study, a moment of 3.30 B.M was observed for the complex, which was indicative of spin equilibrium between the high spin and low spin octahedral geometry^[18].

Table 2: Relevant Infrared and electronic spectra data of the complexes.

Compound	ν(C=O)	ν(C=N)	ν(M-O)	ν(M-Cl)	Electronic spectra (kK)
HL	1733(s)	1649(s) 1581(s) 1546(s)	-	-	29.41
HL ¹	1666(s)	-	-	-	27.32 29.24
Mn(HL)(HL ¹)Cl ₂ .H ₂ O	1733(s) 1622(s)	1649(s) 1580(s) 1547(s)	486(s) 409(s) 474(s)	386(s) 370(s) 353(s)	11.67 22.37 29.76
[Fe(HL)(HL ¹)SO ₄]	1732(s) 1622(s)	1648(s) 1581(s) 1546(s)	486(m) 448(s) 409(m)	386(s) 378(s) 369(s) 358(s)	18.73 24.39 27.40 29.33(sh)

[Co(HL)(HL¹).Cl₂].2H₂O	1733(s)	1649(s) 1581(s)	486(s) 474(m)	386(s) 370(s)	14.88 25.32
	1622(s)	1546(s)	448(s)	356(s)	29.67 48.31
[Ni(HL)(HL¹)Cl₂].H₂O	1733(b)	169(s) 1581(s)	486(s) 474(m)	386(s) 370(s)	14.95 24.94
	1622(s)	1546(s)	448(s) 409(s)	356(s)	29.33
[Cu(HL)(HL¹)Cl₂].H₂O	1732(s)	168(s) 1581(s)	485(s) 474(s)	380(s) 371(s)	16.47 24.94(sh)
	1621(s)	1549(s)	448(s) 409(s)	354(s)	29.64
[Zn(HL)(HL¹)SO₄]	1732(s)	1649(s) 1581(s)	486(s) 448(s)	386(s) 371(s)	14.97 24.94
	1621(s)	1547(s)	409(s)	354(s)	28.90

HL = Riboflavin, HL¹ = 4aminobenzoic acid, s = strong, m = medium, b = broad, w = weak, kK = 1000 cm⁻¹

Similarly, Co(II) complex had two absorption bands at 14.88 kK and 25.32 kK typical of high spin and low spin octahedral geometry assigned to ${}^4T_{1g}(F) \rightarrow {}^4A_{2g}(F)$ and ${}^2E_g \rightarrow {}^2T_{1g}$ transition respectively. An observed moment of 4.09 B.M was corroborated of spin equilibrium between the high spin and low spin octahedral geometry since moments in the range 4.7-5.2 B.M and 1.9-2.4 B. M were reported for high spin and low octahedral complexes ^[19]. The Ni(II) complex showed two absorption bands at 14.95 kK and 24.94 kK typical of 6-coordinate high spin and low spin octahedral geometry. These were assigned as ${}^3A_{2g} \rightarrow {}^3T_{2g}$ and ${}^1B_{2g} \rightarrow {}^1B_{1g}$ transitions respectively, since moment in the range 0.5- 2.7 B.M were reported for Ni(II) complexes with spin-equilibria. The Ni(II) complex gave a moment of 2.58 B.M corroborating spin equilibrium between the high spin and low spin octahedral geometry ^[20]. The Cu(II) complex displayed two absorption bands at 16.47kK and 24.94kK, typical of a 6-coordinate, tetragonal (octahedral) geometry and were assigned to ${}^2B_{1g} \rightarrow {}^2B_{2g}$ and ${}^2B_{1g} \rightarrow {}^2E_g$ transitions. An observed moment of 2.27 B.M was complimentary of this geometry since moments in the range 1.9-2.3 B.M were reported for mononuclear Cu(II) complexes regardless of geometry ^[21,22]. The spectra of Zn(II) complex had only charge transfer transitions from metal to ligand at 14.97 kK and 24.94 kK as no d-d transition was expected. The complex was expected to be diamagnetic because of its d¹⁰ configuration. However, a moment of 0.80 B.M was observed due to impurities ^[7, 23]. In all cases, electronic spectra and magnetic moments corroborated octahedral geometry for the metal(II) complexes (Figure 1).

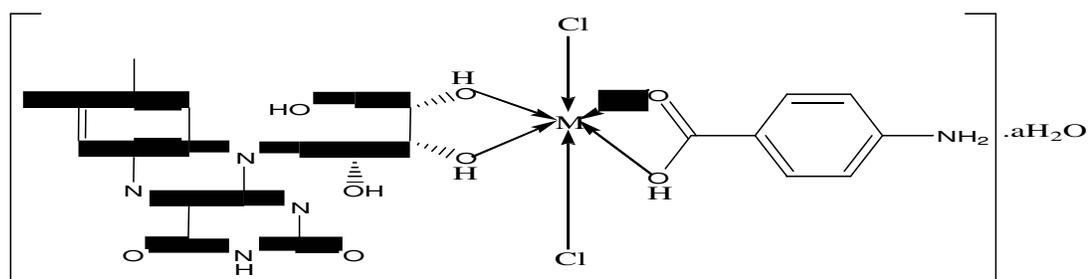


Figure 1: Proposed structure for metal(II) complexes (M = Mn, Co, Ni, Cu; a = 0.5).

Infrared spectroscopy

The relative infrared spectral data are presented in Table 2. The bands at 3496 cm⁻¹ and 3495 cm⁻¹ in Riboflavin(HL) and 4-aminobenzoic acid(HL¹) were assigned as $\nu(N-H)$ stretching vibrations ^[7,24]. In the metal(II) complexes, these bands remained un-shifted indicative of non-coordination of the imine nitrogen atom of the Riboflavin(HL) and the amine nitrogen atom of the 4-aminobenzoic acid(HL¹) to the metal(II) ions respectively. The strong vibration bands at 1733 cm⁻¹ and 1666 cm⁻¹ in Riboflavin(HL) and 4-aminobenzoic acid(HL¹) were assigned as $\nu(C=O)$ stretching vibration. In Riboflavin, the band was un-coordinated with the carbonyl oxygen whereas in 4-aminobenzoic acid (HL¹), $\nu(C=O)$ band shifted to the range 1621-1622 cm⁻¹ in the metal(II) complexes respectively. This confirmed the non-coordination of Riboflavin (HL) via carbonyl oxygen atom and coordination for 4-aminobenzoic acid (HL¹) through carbonyl oxygen atom ^[11]. The azomethine, $\nu(C=N)$ stretching vibration at 1649-1546 cm⁻¹ in Riboflavin(HL) remained un-shifted in the metal(II) complexes indicative of non-coordination of the imine nitrogen to the metal(II) ions. The broad bands at 3377 cm⁻¹ and 3382 cm⁻¹ in Riboflavin (HL) and 4-aminobenzoic acid (HL¹) respectively were assigned as $\nu(O-H)$. These bands were shifted to 3370-3334 cm⁻¹ in the metal(II) complexes, indicative of coordination of the hydroxy oxygen atoms in the Riboflavin(HL) and 4-aminobenzoic acid(HL¹) to the metal ions respectively. The new bands in the range 409-486 cm⁻¹ and 353-386 cm⁻¹ were assigned to $\nu(M-O)$ and $\nu(M-Cl)$ respectively. Conversely, these bands were absent in the spectra of the Riboflavin (HL) and 4-aminobenzoic acid(HL¹), confirming coordination in the metal(II) complexes ^[11,25].

Antimicrobial activities

The results of antimicrobial activities of metal(II) complexes of Riboflavin and 4-aminobenzoic acid against different clinical, food, and environmental strains of organisms are presented in Table 3. None of the metal(II) complexes, ligands and the positive standard (Streptomycin) was active against *E. coli* (typed strain) with the exception of Riboflavin with an inhibitory zone of 7.0 mm. Similarly, the metal(II) complexes and ligands were inactive against *Streptococcus spp*, *Staphylococcus sp*, *Pseudomonas sp* (environmental strain), *Bacillus sp* (environmental strain) and *E. coli* (clinical strain) respectively. However, Riboflavin was active against *Streptococcus pyogenes* with inhibitory zone of 7.0 mm, and 4-aminobenzoic acid was active against *Streptococcus sp* (Blood) with inhibitory zone of 10.0 mm respectively. The inactivity of some of the metal(II) complexes were attributed to their probable lipophobic nature and as such could not penetrate through the lipid membrane of the bacteria [26,27]. Contrarily, all the metal(II) complexes, their ligands and positive standard were active against *Pseudomonas sp* (Clinical Isolate) with inhibitory zones range of 7.0-15.0 mm. The Zn(II) complex and Streptomycin were inactive against *Bacillus sp* (Food Strain) while the remaining metal complexes and ligands were active against this bacteria with inhibitory zones range of 5.0-8.0 mm. It's interesting to note that environmental strain of *Bacillus sp* and *Pseudomonas sp* were resistant to the metal complexes, Riboflavin and Benzoic acid whereas the food strain of *Bacillus sp* and clinical strains of *Pseudomonas sp* had activity against all the metal(II) complexes, with the exception of the Zn(II) complex for the former bacteria, Riboflavin and Benzoic acid. These observations were attributed to development of efflux pump by the bacteria or the useage of the metal complexes as terminal electron acceptors in anaerobic respiration by these bacteria [28]. Similarly, Riboflavin, HL, had antimicrobial activity against *E. coli* (Type strain) but not *E. coli* (Clinical strain) due to antibiotic resistance of clinical organisms [29]. Expectedly, Streptomycin had better activity than the metal(II) complexes and ligands with inhibitory zones range of 9.0-29.0 mm.

Table 3: Antimicrobial activities of the Riboflavin and 4-aminobenzoic acid and their complexes.

Complex organism	[Mn(HL)]	[Fe(HL)]	[Co(HL)]	[Ni(HL)]	Cu(HL)	Zn(HL)	Streptomycin Sulphate
	H ₂ O	H ₂ O	Cl ₂ .1.2H ₂ O	H ₂ O					
<i>E.coli</i> ^T <i>E.coli</i>	R	R	R	R	R	R	7.0±0	R	R
<i>Bacillus sp</i> ^C (Food)	R	R	R	R	R	R	R	R	15.0±0
<i>Bacillus sp</i> ^E	8.0±0	7.0±0	5.0±0	7.0±0	7.0±0	R	8.0±0	7.0±0	R
<i>Candida albicans</i>	R	R	R	R	R	R	R	R	29.0±0
<i>Proteus mirabilis</i>	R	R	9.0±0	7.0±0	R	R	9.0±0	R	20.0±0
<i>Salmonella sp</i>	10.0±0	7.0±0	R	R	R	R	R	R	9.0±0
<i>Staphylococcus sp</i>	R	R	7.0±0	7.0±0	R	R	7.0±0	R	R
<i>S. pyogenes</i>	R	R	R	R	R	R	R	R	21±0
<i>Streptococcus sp</i> ^B	R	R	R	R	R	R	7.0±0	R	13.0±0
<i>Pseudomonas sp</i> ^C	R	R	R	R	R	R	R	10.0±0	12.0±0
<i>Pseudomonas sp</i> ^E	9.0±0	8.0±0	7.0±0	7.0±0	9.0±0	9.0±0	7.0±0	11.0±0	15.0±0
	R	R	R	R	R	R	R	R	16.0±0

HL = Riboflavin; HL¹ = 4-aminobenzoic acid; R = Resistance; C = Clinical Isolate; E= Environmental Strain; T = Typed strain; B = Blood; F = Food strain

Ferrous metal ion chelating activity of metal (II) complexes

The activities of metal ions in biology systems such as Vitamin B12 and Hemoglobin have stirred up renewed interest in the development of metal-based therapy because the former is used to treat cyanide poisoning, and allergy [30], while the latter in mesangial cells of the kidney function as an antioxidant and a regulator of iron metabolism [31]. Thus, it is necessary to mimic these biomolecules *in-vitro* and investigate their effectiveness as chemotherapeutic agents against infectious diseases and various carcinomas. Transition metal such as ferrous ion can facilitate the production of reactive oxygen species (ROS) within animal and human systems, and thus, the ability of substances to chelate metal ion can be a valuable antioxidant capability [32]. Iron, in nature, can be found as either ferrous (Fe²⁺) or ferric ion (Fe³⁺), with the latter form predominating in foods. Transition metals such as Fe²⁺, Cu⁺, Co²⁺ may serve as catalysts for the initial formation of free radicals. Free radical induced lipid oxidation in human

bodies is related to the pathological process of many diseases, such as cardiovascular disease^[33, 34], some cancerous disorders and diabetics^[35]. Furthermore, it is well known that polyphenol compounds such as Riboflavin are very good chelating agents having the ability to stabilize transition metals in living systems and inhibit generation of free radicals. In this study, the antioxidant studies on mixed –ligand, Riboflavin and Benzoic acid metal(II) complexes showed that the Zn(II) complex exhibited maximum inhibitory value of 61.66% while the Fe(II) complex had the minimum inhibitory value of 19.12%. Interestingly, the Ni(II) complex had about the same inhibitory activity (~33%) as the standards-ascorbic acid and α -tocopherol. Thus, the metal(II) complexes are potential anticancer agents (Table 4).

Table 4: Antioxidant data of the mixed ligand metal(II) complexes.

Complexes	Absorbance (error)	% Inhibition(error)
[Mn(HL)(HL ¹)Cl ₂].H ₂ O	0.672(0.0030)	54.28(0.1790)
[Fe(HL)(HL ¹)SO ₄]	0.237(0.0023)	19.12(0.1617)
[Co(HL)(HL ¹)Cl ₂].2H ₂ O	0.533(0.0022)	43.05(0.1554)
[Ni(HL)(HL ¹)Cl ₂].H ₂ O	0.406(0.0023)	32.77(0.1386)
[Cu(HL)(HL ¹)Cl ₂].H ₂ O	0.271(0.0029)	21.91(0.3060)
[Zn(HL)(HL ¹)SO ₄]	0.763(0.0055)	61.66(0.3579)
A ₁	0.413(0.0010)	33.36(0.0464)
α_1	0.411(0.0021)	33.19(0.1722)

HL = Riboflavin, HL¹ = 4-aminobenzoic acid, A₁ = ascorbic acid, α_1 = α -tocopherol

CONCLUSION

From the infrared spectra data, the ligands behaved as bidentate and were coordinated to the metal ion through the oxygen atoms of hydroxy groups in Riboflavin and the carboxylate oxygen atoms of the 4-aminobenzoic acid respectively. Electronic spectra and room temperature magnetic moments corroborated octahedral geometry for all the metal(II) complexes. Furthermore, the antimicrobial studies showed that the metal(II) complexes had mostly lower activity in comparison to Streptomycin that was active against almost all the organisms with the exception of *E. coli* (Typed strain), *Bacillus sp* (Food Strain) and *Salmonella sp*. In addition, the antioxidant activity of the metal(II) complexes showed that the Zn(II) complex had the best activity being about twice that of the standards, ascorbic acid and α_1 -tocopherol.

REFERENCES

- Halli MB, Patil VB, Sumathis RB, Mallikarjini K. Synthesis, characterization and biological activity of mixed-ligand metal(II) Complexes derived from Benzofuran-2-carbohydrazide Schiff base and malonyldihydrazide. *Der pharma chemical*. 2012; 4: 2360-2367.
- Moustafa ME. Synthesis, structure and biological activity studies on some lanthanide chelates with O-and N- containing ligands. *Spectroscopy Lett*. 2005;38: 23-34.
- Ogundiran KO, Ajanaku KO, James OO, Ajani OO, Nwinyi CO, et al. Fe(II) and Co(II) complexes of mixed antibiotics: Synthesis, characterization, antimicrobial potential and their effect on alkaline phosphatase activities of selected rat tissues. *Int J Pharm Sci*. 2008;3: 177-182.
- Osoyole AA, Ekennia AC, Achugbu BO, EtukGH. Synthesis, spectroscopic characterisation and structure related antibacterial activities of some metal(II) complexes of substituted trifluorobutanol. *Elixir Appl Chem*. 2013;59:15848-52.
- Osoyole AA, Ekennia AC, Achugbu BO. Synthesis, Spectroscopic characterization and antibacterial Properties of some Metal (II) Complexes of 2-(6-methoxybenzothiazol-2-ylimino)methyl-4-nitrophenol. *Research & Reviews: Journal of Pharmaceutical Analysis*. 2013;2: 1-5
- Osoyole AA, Ekennia AE, Ozukwe AE. Synthesis, spectroscopies and antibacterial properties of some metal(II) mixed ligand complexes of Riboflavin and 2,2'-bipyridine. *Research & Review: Journal of Chemistry*. 2014;2322-0082.
- Osoyole AA, OBA Agbaje, BO Ojo. Synthesis, characterization and antibacterial properties of some Heteroleptic metal (II) complexes of paracetamol and vanillin. *Asian J Pharm and Clinical Res*. 2014;7:145-149
- Shivankar VS, Thakkar NV. Synthesis, characterization and antimicrobial activity of some mixed ligand Co(II) and Ni(II) complexes. *ActaPoloniaePharmaceutica*. 2003;60: 45-50.
- Vogel C, Grill S, Schilling O, Stülke J, Mack M, et al. Characterization of riboflavin (vitamin B2) transport proteins from *Bacillus subtilis* and *Corynebacterium glutamicum*. *Journal of bacteriology*. 2007;189: 7367-7375.
- Barker BM, Bender DA (Eds.) *Vitamins in medicine*. William Heinemann Medical Books, London, 1995.

11. Chadar SN, Khan F. Electrode Kinetics and Ternary Complexes of [MnII-antibiotics-vitamins B₂] vis-à-vis kinetics of electrode Reaction. *J Indian chem Soc.* 2006;83: 1242-1247.
12. Escott-stump S (eds) *Nutrition and Diagnosis-Related Care.* (6thedn), Pa: Lippincoli Williams and Wilkins, Philadelphia, 2008.
13. Powers HJ. Riboflavin (vitamin B-2) and health. *The American journal of clinical nutrition.* 2003;77: 1352-1360.
14. Roberts SB, Morrow FD, Evans WJ, Sheppard DC, Dallal GE, et al. Use of p-aminobenzoic acid to monitor compliance with prescribed dietary regimens during metabolic balance studies in man *Am J Clin Nutr.* 1990; 51: 485-488.
15. de Souza R F, De Giovani W F. Antioxidant properties of complexes of flavonoids with metal ions. *Redox Report.* 2004; 9: 97-104.
16. Ekennia AC, Onwudiwe DC, Osowole AA. Synthesis, Spectral, thermal stability and antibacterial studies of copper, nickel and cobalt complexes of *N*-methyl-*N*-phenyl dithiocarbamate. *Journal of Sulfur Chemistry.* 2015; 36: 96-104.
17. Saha A, Majumda P, Goswami S. Low spin Mn(II) and Co(II) complexes of *N*-aryl-2-pyridylazophenylamines; New tridentate *N,N,N*-donors derived from cobalt mediated aromatic ring amination of 2-(phenylazo)pyridine. Crystal structure of manganese(II) complex. *J ChemSoc (Dalton Transactions).* 2000; 11: 1703-1708.
18. Olguín J, Brooker S. Spin crossover active iron(II) complexes of selected pyrazole-pyridine/pyrazine ligands. *Coordination Chemistry Reviews.* 2011;255: 203–240.
19. Goodwin HA. Spin Crossover in Cobalt(II) Systems. *Top Curr Chem.* 2004; 234: 23-27.
20. Ohtsu H, Tanaka K. Equilibrium of low- and high-spin states of Ni(II) complexes controlled by the donor ability of the bidentate ligands. *Inorganic Chemistry.* 2004;43: 3024-3030.
21. Katsarou ME, Elani K, Efthimiadou GP, Karaliota A, Vourloumis D. Novel copper(II) complex of *N*-propyl-norfloxacin and 1,10-phenanthroline with enhanced antileukemic and DNA nuclease activities. *Journal of medicinal chemistry.* 2008;51(3): 470-478.
22. Agwara MO, Ndifon PT, Ndosir NB, Paboudam AG, Yufunji DM, et al. Synthesis, Characterization and antimicrobial activities of Co(II), Cu(II) and Zn(II) mixed-ligand complexes containing 1,10-phenanthroline and 2,2'-bipyridine. *Bull chemSoc Ethiop.* 2010;24: 383-9.
23. Raman N, Ravichandran S, Thangaraja C. Cu(II), Co(II), Ni(II) and Zn(II) complexes of Schiff base derived from benzil-2,4-dinitrophenyl hydrazone with aniline. *J ChemSoc (Indian Academic Society).* 2004;4: 116– 215.
24. Ajibade PA, Idemudia OG. Synthesis, characterization and antibacterial studies of Pd(II) and Pt(II) complexes of some diaminopyrimidine derivatives. *Bioinorganic Chem Appl.* 2013;1-8.
25. Dinoiu V, Voicescu M, Lungu L, Savoiu M, Vasilescu M. Spectroscopic Study on the Riboflavin – Plant Extracts Interaction. *Rev Chim (Bucharest).* 2011;62: 1111-1114.
26. Matangi S, Pragathi J, Bathini U, Gyana K. Synthesis, Characterization and antimicrobial studies of transition metal complexes of Schiff base ligand derived from 3-ethoxy salicylaldehyde and 2-(2-aminophenyl)-1,11-benzimidazole. *E J Chem.* 2012;9: 2516-23.
27. Nasir R, Shah A, Baluja S, Chanda S. Synthesis and antimicrobial activity of some Schiff base complexes. *J of the Serbian society.* 2006;71: 733-44.
28. Gadd, GM. In *Microbial Mineral Recovery* (Ehrlich, HL and Brierley, CL., Eds.) McGraw Hill, New York. 1990;249-275.
29. Wang M, Tran J H, Jacoby G A, Zhang Y, Wang F, et al. Plasmid-Mediated Quinolone Resistance in Clinical Isolates of *Escherichia coli* from Shanghai, China. *Antimicrob Agents Chemother.* 2003;47: 2242–2248.
30. Dart R C Hydroxocobalamin for acute cyanide poisoning: new data from preclinical and clinical studies; new results from the prehospital emergency setting. *Clinical Toxicology.* 2006;44: 1–3.
31. Biagjoli M, Pinto M, Cesselli D, et al. Unexpected expression of alpha- and beta-globin in mesencephalic dopaminergic neurons and glial cells. *Proc Natl Acad Sci USA.* 2009, 106: 15454–15459.
32. Halliwell B, Gutteridge JMC. Oxygen toxicology, oxygen radicals, transition metals and disease. *Biochem J.* 1984;219: 1-4.
33. Miller ER, Appel LJ, Levander OA, Levine DM. The effect of antioxidant vitamin supplementation on traditional cardiovascular risk factors. *J cardiovascular risk.* 1997;4: 19-24.
34. Joshipura KJ, Hu FB, Manson JE, Stampfer MJ, Rimm EB, et al. The effect of fruit and vegetable intake on risk for coronary heart disease. *Ann of intern med.* 2001;134: 1106-1114.
35. Holick MF Vitamin D: importance in the prevention of cancers, type 1 diabetes, heart disease, and osteoporosis. *Am J clinnutri.* 2004;79: 362-71.