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Synthesis, Spectral Characterization and *In Vitro* Antihelmintic Evaluations of Cu(II) and Co(II) Complexes with A Novel Schiff Base Ligand

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Abstract

The novel Schiff base ligand (Z)-2-(benzalideneamino)-3-(1H-indol-3-yl)propanoic acid was synthesized by condensation reaction of benzaldehyde and tryptophan. The complexes of this ligand of Cu (II) and Co (II) were successfully synthesized and characterized physiochemically and spectroscopically. The ligand and the complexes showed a good yield ranging from 95 % - 98 %. The FTIR spectroscopic data showed appearance of the azomethine (C=N), carbonyl group (C=O) and -OH group with peak at 1692 cm^{-1}, 1580 $cm^{-1}$and 3399 cm^{-1} respectively in the spectrum of Schiff base. The molar conductivity data of the Schiff base was found to be 3.5 $\Omega^{-1}cm^2mol^{-1}$ indicating non-electrolytic nature whereas that of Cu and Co complexes ranges between 103 -160 $\Omega^{-1}cm^2mol^{-1}$ indicated that the complexes were weak electrolyte in water. On the basis of magnetic and electronic spectral data, octahedral geometry was proposed for all the complexes. The UV-Vis spectra showed metal-ligand bond formation. The complexes demonstrated various significant antihelmintic activities against *Taenia spp* compared to the ligand. The Co complex showed similar biological activity compared to the ligand while Cu complex showed biological activity higher than the ligand. Both the Co complex and the ligand showed a lower activity compared to the standard drug except for Cu complex that showed the same activity as the standard drug hence, the Cu complex synthesized is a good replacement for the existing drug.

Keywords: Antihelmintic, Schiff base, percentage mortality and (Z)-2-(benzalideneamino)-3-(1H-indol-3-yl)propanoic acid.

Introduction

Schiff base coordination chemistry has long been a topic of intense study. Schiff bases have been employed for the synthesis of metal complexes because of their strong metal binding capacity and ease of production. They can form coordination bonds with a variety of metal ions through azomethine and phenolic groups [1]. The creation of Schiff base complexes with unique structural characteristics and peculiar physiochemical characteristics is a significant biological process and a current area of study in coordination chemistry. Its remarkable therapeutic uses are marketed as medications for the treatment of cancer, angiogenesis, Alzheimer's disease, inflammation, and the inhibition of bacterial, fungal, and microbiological activity [2].

In a similar process, 4-Chloro-2-oxo-2H-Chromenene-3-carbaldehyde was reacted with different anilines in rectified spirit as solvent to yield a series of the title compounds i.e. 4-chloro-3-((substituted-phenylimino)methyl)-2H-chromene-2-one. These compounds were characterized on the basis of their spectral(IR,^{1}H NMR) data and evaluated for antimicrobial activity *in vitro* against gram negative bacteria and fungi. The Compound was found to be most active with an MIC of 15I µg/mL against all the tested organisms [3].

Several new thiazolyl/thiazolinyl /benzothiazolyl Schiff bases have been Designed, synthesized and identified . The compounds have been reported to act as lipoxygenase inhibitors affecting inflammation and or psoriasis. The compounds were screened for their reducing activity (with stable free radical 1,1-diphenyl-2-picrylhydrazyl,DPPH) and for inhibition of soybeans lipoxygenase (LOX).Anti-inflammatory activity was examined *in vivo* using the paw edema method. The synthesis of twenty new Schiff bases derived from ortho hydroxyl aldehydes and aromatic with high yields via condensation in ethanol in the presence of catalytic amounts of sulfuric acid have been reported [5]. These azomethines, useful as ligands, intermediates in organic synthesis or potential biologically active substances, have been characterized by elemental analysis, IR, ^{1}H and ^{13}C NMR spectra.

Schiff base complexes have a wide range of biological properties; antitumor, antiviral, antifungal, antibacterial, nematicidal etc [6]. Schiff bases are used in the treatment of diabetes, cancer and many other diseases in the body [7]. As biological models, they help in understanding the structure of biomolecules and biological processes occurring in living systems [6]. They assist among others in photosynthesis and oxygen transport in organisms [8]. The chemistry of Schiff base transition metal complexes is largely appealing because of their broad profile pharmacological activity providing a variety of compounds with different activities [5], [9]. Transition metal complexes of Schiff base ligands have gained considerable attention recently due to their antibacterial activities in addition to their

spectroscopic properties [10]. With rising mortality from infectious diseases directly linked to bacteria exhibiting multiple antibiotic resistance due to ineffective treatment, the discovery of novel antibacterial medicines with more effective mechanisms of action is unquestionably urgently needed [11]. Schiff base transition metal complexes appear to be a future medical candidate to select compounds as therapeutic agents [12] due to their wide range of antibacterial activities compared to the parent Schiff base[13], [14]. Kumar *et al*., [15] synthesized Co (II) and Cu (II) complexes of the Schiff base derived from vanillin and acetoacetanilide with ethylenediamine and characterized based on their elemental analysis, conductivity measurements, IR and electronic spectral, HNMR. The antimicrobial study was carried with the ligands and its metal complexes. The result revealed that the Schiff base transition metal complexes showed significant activity against some fungi and bacteria than the uncomplexed ligands. Azomethine compounds have shown wide range of biological activities such as antimalarial, anticancer, antibacterial [16], anti-inflammatory [2], antifungal, antibacterial, antileishmania, antiviral, antifertility and antipyretic activities [17] with the nematicidal activity reported in literature [18].

Research findings by some authours [2], [19] and [20] in the areas of interaction of metal ions with organic ligands have all shown that the complexes possess better antimicrobial activity compared to free liquids.

The biology activities of Cu(II) and Co(II) Complexes have been studied using different Schiff base ligands as published in some literatures [1],[21] and other available publications indicating that there is dearth of literature as regard studies bordering on synthesis and characterization of (Z)-2-(benzylideneamino)-3-(1H-indo-3-yl)propanoic acid or benzalidenetryptophan (BAT) and its complexes of Cu(II) and Co(II) with their antihelmintic profiles. Furthermore, there are reports of government regulation on some available drugs hence the need to synthesize alternative drugs.

Material and Methods

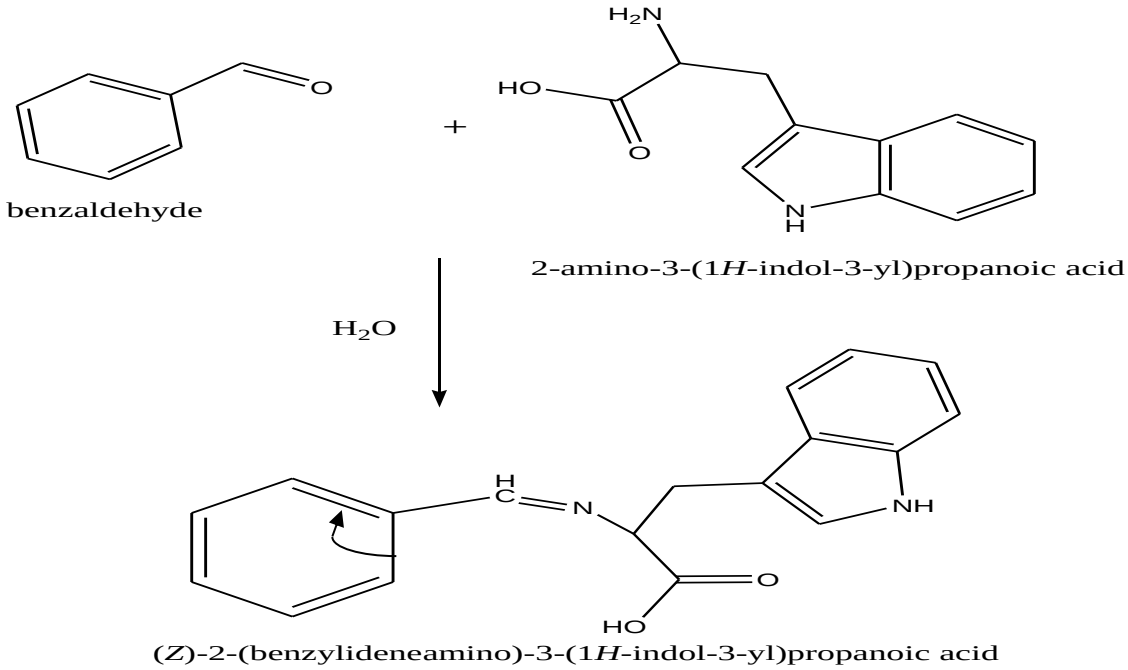
Reagents: Copper (II) chloride, Zinc (II) chloride, Cobalt (II)chloride, Cadmium (II) Chloride, distilled water, benzaldehyde, tryptophan and among others are the reagents used for this study. All the reagents used are of analytical grades and purchase from BDH and Merck chemical company hence, no further purification was needed as a result of their high percentage purity.

Apparatus: routine laboratory apparatus /equipments used include conical flask, spatula, filter papers, funnels, pipette, petri dish, beakers, measuring cylinders, thermometer, conductivity meters, *Shimadzu* infrared spectrophotometer, *Shimadzu* 1800 UV-visible double beam PC scanning spectrophotometer, heat stunner PW184 weighing balance, 98-1-B and temperature regulating mantle. The IR (Cary 20, and EDXRF

Unicam 929) characterization of the Schiff base and the complexes were carried out at the Umaru Musa Yar'adua University Katsina State Nigeria Central Research Laboratory while the biological study was done in the Microbiology Department of Benue State University Teaching hospital Makurdi Nigeria.

Preparation of ligand

The Schiff base (Z)-2-(benzylideneamino)-3-(1H-indo-3-yl)propanoic acid (BAT) was synthesized by measuring l mL (0.01mol/dm^3) of benzaldehyde and mixed with 1g (0.005mol/dm^3) tryptophan crushed in a crucible and both were stirred in a 250 mL beaker continuously for about 5 minutes until a bright yellow crystals with 99.6% yield formed, air-dried for 7 days and preserved in an air-tight bottle to avoid further reaction [3]. The Schematic representation of the synthesis of the ligand is as shown in Figure 1 below.



Preparation of metal complexes:

Exactly 1g of $CoCl_2$ dissolved in 5 mL solvent (Distilled and deionized water) at ambient temperature. 2g of the Schiff base ligand dissolved in 10mL solvent (Distilled and deionized water at 35 ^{0}C). 1 mL of aqueous solution of the ligand was added to 5 mL $CoCl_2$ aqueous solution and mixed thoroughly. This was repeated four times and mixed in the constant 5mL metal solution and stirred in a 250 mL beaker until crystals were formed. The crystals formed were air-dried for seven (7) days. This procedure was repeated for the preparation of

Cu(II) complex [2] The metals complexes were formed by the reaction between the metal salts (MCl_2) and the ligand with 1:2 stoichiometry as shown in Equation 1. The structure of the complex is presented in Figure 2 below.

$$MCl_2 + 2\,C_{18}H_{16}N_2O_2 \xrightarrow{30\text{-}32\ ^\circ C} [M(C_{18}H_{16}N_2O_2)_2]Cl_2 \quad (1)$$

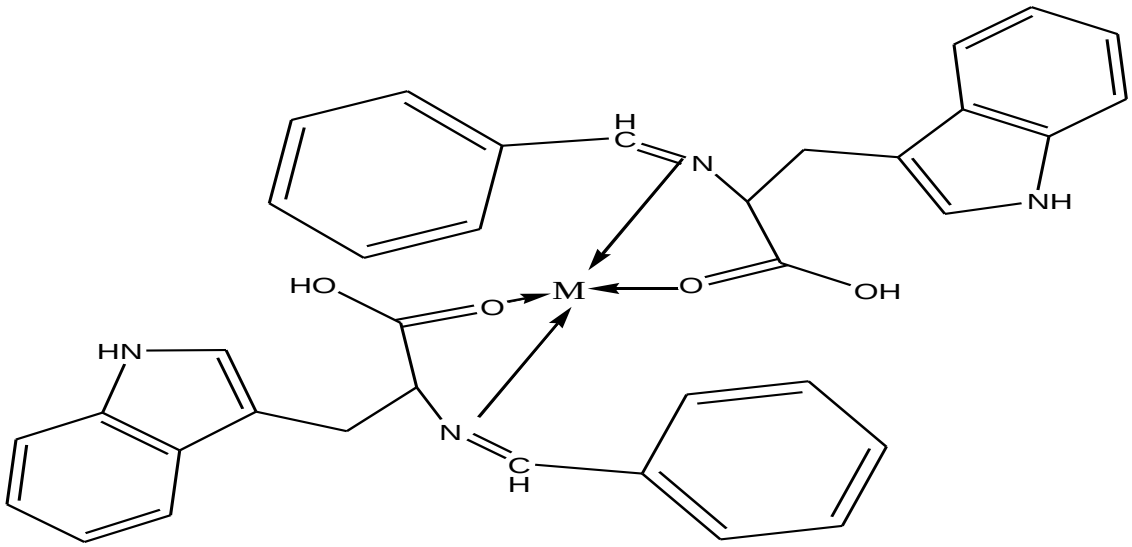


Figure 2: The proposed structure of the complexes

Where **M**= Co(II) and Cu(II)

Results and Discussion

The physicochemical properties of the ligand and complexes are presented in Table 1 below. All the complexes precipitated as powders, coloured, stable and hygroscopic in nature. The complexes were slightly soluble in water and ethanol. The elemental analysis showed that the complexes have 1:2 stoichiometry of the type $[M(C_{18}H_{16}N_2O_2)_2Cl_2]$.

Table1: Physiochemical Properties of the Ligand and its Metal Complexes

Compound	Molecula formula	Solubility		Molar conductance $\Omega^{-1}cm^2mol^{-1}$	Melting point (°C)	Colour	Yield (%)
		Water	**Ethanol**				
BAT	$C_{18}H_{16}N_2O_2$	SS	NS	3.5	290-294	Yellow	98
$Co(L_2)$	$Co(C_{18}H_{16}N_2O_2)_2$	SS	SS	160	66-71	Orange	92
$Cu(L_2)$	$Cu(C_{18}H_{16}N_2O_2)_2$	SS	SS	103	67-281	Dark blue	95

SS= Sparingly Soluble ; NS= Not Soluble

The conductance values, which are presented in the Table 1, indicate that the complexes are weak electrolyte while the ligand is non-ionic [21]. The melting points of complexes were determined and found to be within the range of 66-281(°C). The solubility test shows that the ligand and both complexes are slightly soluble in water while only the ligand was not soluble in ethanol.

The EDXRF analysis to determine the metallic percentage composition of the ligand and the complexes was carried out and shown as Table 2 and their spectra presented in Figures 3, 4 and 5. The results show the percentage composition of Cu and Co as 0.023 and 0, 0.123 respectively in the ligand sample. This indicates little or no metal and hence, no formation of coordination bond between the ligand and the metal ions, this is in agreement with result obtained elsewhere [4]. The result of the metallic composition conducted on the Cu complex gives the following data: Cu=3.07 and Co=0. This result indicates the presence of Cu and probably Cu-ligand bond formation. Similarly, the result obtained from the analysis of Co complex show that Cu=0.010 and Co=19.4 confirming the presence of Co and Co-ligand bond formation.

Table 2: The Elemental Analysis of the Ligand and Metal Complexes

Compound	**Percentage composition**	
	Cu	**Co**
Ligand	0.02265	--
Cu complex	3.0741	--
Co complex	0.0097	19.423

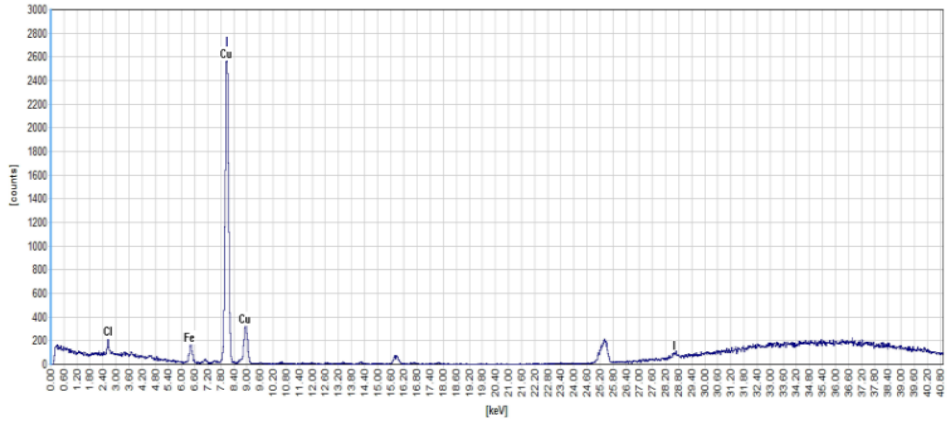


Figure 3: EDXRF for Cu Complex

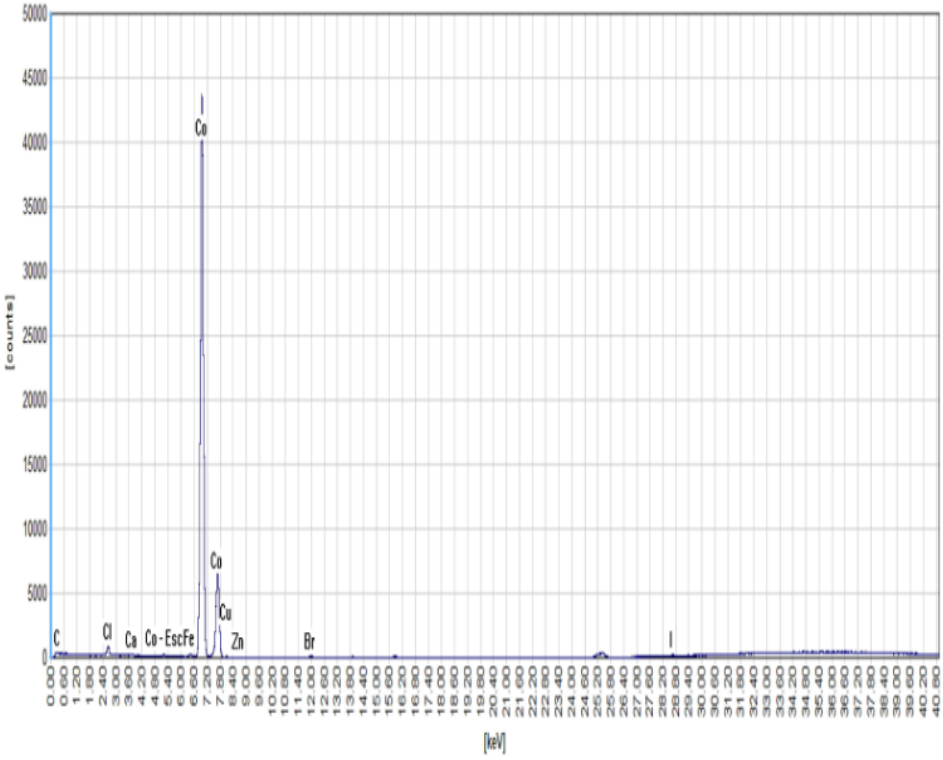


Figure 4: EDXRF for Co Complex

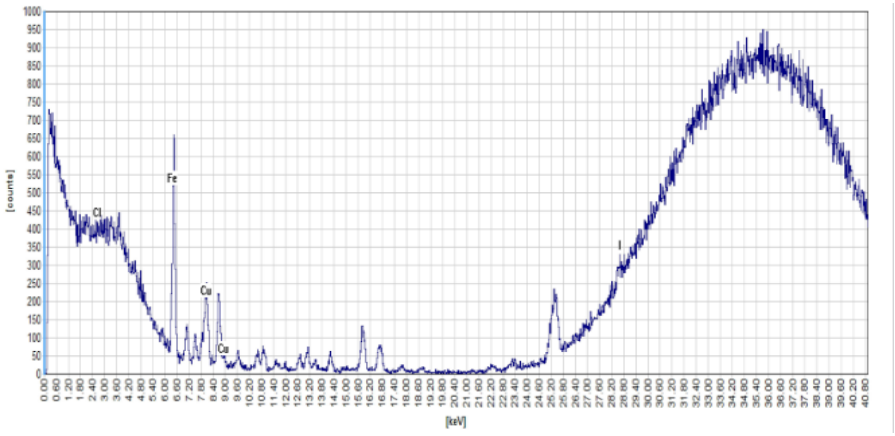


Figure 5: EDXRF for the Ligand

Table 3: Electronic Spectra and Magnetic Data of the Ligand and Complexes

Ligand/Complexes	Magnetic Moment (BM) μ_{eff}	Vibrational frequency(cm^{-1})	Assignment	Geometry
BAT	-	22300	charge transfer	
				octahedral
Co(II)complex	4.65	8467	$^4T_1g(F) \rightarrow {}^4A_2g(F)$ (V_1)	
		16304	$^4T_1g(F) \rightarrow {}^4A_2g(F)$ (V_2)	octahedral
		23264	$^4T_1g(F) \rightarrow {}^4T_1g(P)$ (V_2)	
Cu(II)complex	1.73	15200	$^2Eg \rightarrow {}^4T_2g(V_1)$	
		17028	$^2B_1g \rightarrow {}^2A_1g(V_2)$	octahedral

Electronic absorption Spectra and magnetic moment:

The electronic spectral data of the ligand and complexes are represented in Table 2. In the ligands, the band appearing in the 22300cm^{-1} range is assigned to the azomethine chromophore π-π*transition. This report agreed with the study done elsewhere [22].

In the complexes, the azomethine chromophore shift indicates that the imino nitrogen is involved in coordination to the metal ion. The electronic spectra of [$Co(L_2$] gave three absorptions at 8467 cm^{-1},

16304 cm^{-1} and 23264cm^{-1} assigned to $^4T_1g(F) \rightarrow {}^4T_2g\ (F)(V_1)$, $^4T_2g(F) \rightarrow {}^4A_2g(F)(V_2)$ and $^4T_1g(F) \rightarrow 4T_1(P)(V_2)g$ transitions with magnetic moment value 4.65 B.M indicate octahedral cobalt(II) complex [23].

The electronic absorption spectrum of Cu(II) complex showed two bands at 15200cm^{-1} and 17028cm^{-1} corresponding to the transition $^2Eg \rightarrow {}^4T_2g(V_1)$ and $^2B_1g \rightarrow {}^2A_1g(V_2)$ respectively with the magnetic value of 1.73 B.M. This result supports an octahedral geometry with d_x^2-y^2 ground state which agreed with the finding conducted by [24].

From the magnetic susceptibly which confirm the geometry of the metal complexes, it then follows that the chloride ions acted as anionic ligand as shown in Equation 2.

$$MCl_2 + 2C_{18}H_{16}N_2O_2 \xrightarrow{30\text{-}32^{\circ}C} [M(C_{18}H_{16}N_2O_2)_2Cl_2] \quad (2)$$

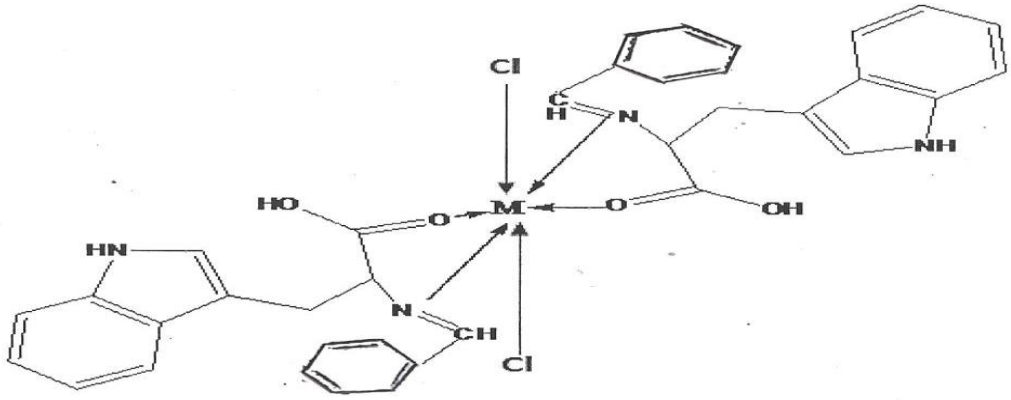


Figure 6: The General Structure of the Synthesized Complexes

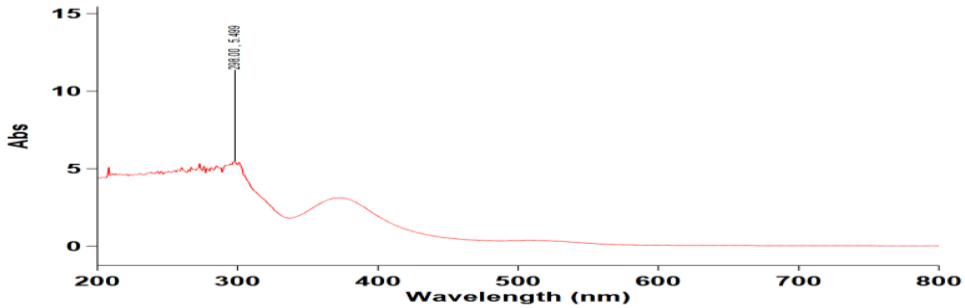


Figure 7: UV Spectrophotometer For Cu Complex

UV Spectra

In the BAT ligand, a sharp band appeared at 303nm corresponding to π-π* transition. The Co complex showed broad band at 370nm indicating a bathochromic shift. This red shift observed in the absorption during the formation of Co complex indicates coordination of the ligand to Co^{2+} ion. Cu complex showed a sharp band at 298nm indicating hypsochromic shift which is an indication of bond formation between the ligand and its metal ion. These observations agreed with already reported literature [21]. The UV spectra for Cu, Co and the ligand are represented as Figure 7-9 respectively.

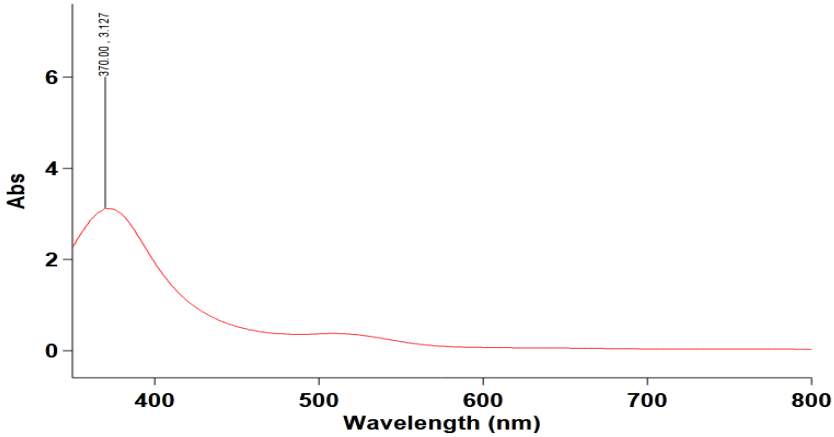


Figure 8: UV Spectrophotometer For Co Complex

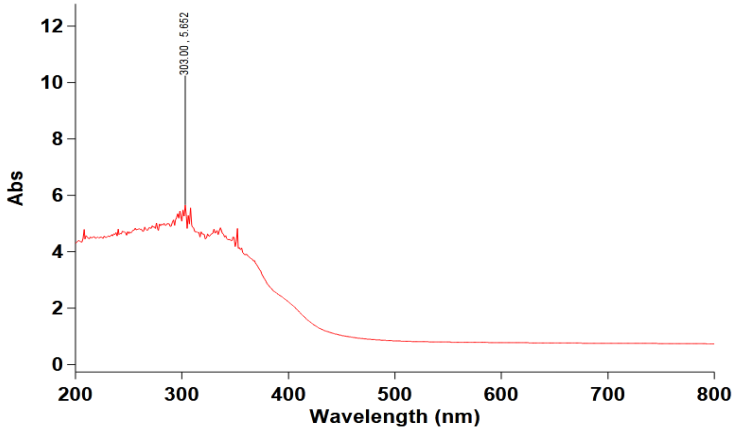


Figure 9: UV Spectrophotometer For BAT

FTIR Characterization

1. The infrared spectral for the ligands and synthesized complexes were recorded using KBr pellets in the range of 4000-450 cm^{-1} and provided valuable information regarding the nature of the functional groups attached to the metal ion. The vibrational frequency data for the ligand L and its corresponding copper (II) and cobalt (II) complexes shown as Table 4 reveal significant insight into the coordination chemistry and bonding interactions within these complexes. The spectra of the ligand and complexes are presented if Figures 10, 11 and 12 below.

Table 4: Some Important Infrared band (cm^{-1}) of the Ligand and its Metal Complexes

Ligand/Complex	C=N	C=O	C=C	NH	OH	M-N	M-O
BAT	1692	1580	1386	3399	3034	-	-
$[Cu(L)_2]$	1684	1560	1453	3399	3026	805	700
$[Co(L)_2]$	1610	-	1401	3369	3175	725	693

For the ligand L, the C=N, C=O, C=C, NH, and -OH stretching frequencies are observed at 1692 cm^{-1}, 1580 cm^{-1}, 1386 cm^{-1}, 3399 cm^{-1}, and 3034 cm^{-1}, respectively. Upon complexation, these frequencies undergo noticeable shifts, indicating coordination through these functional groups.

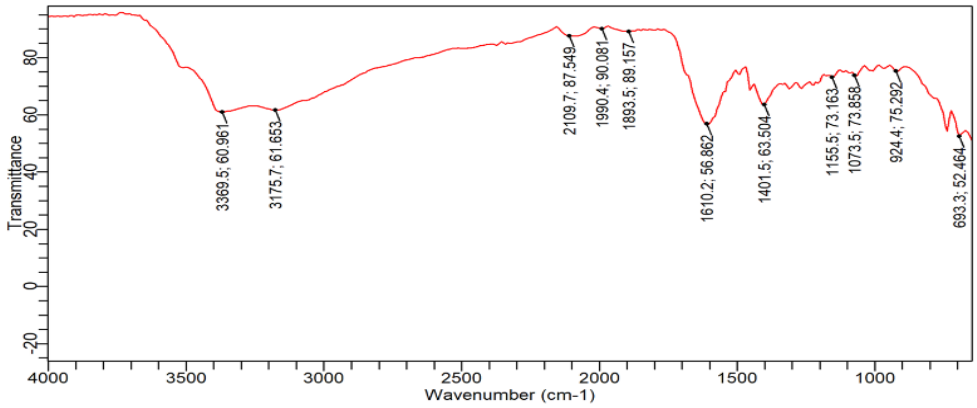


Figure 10: FTIR OF CuL_2

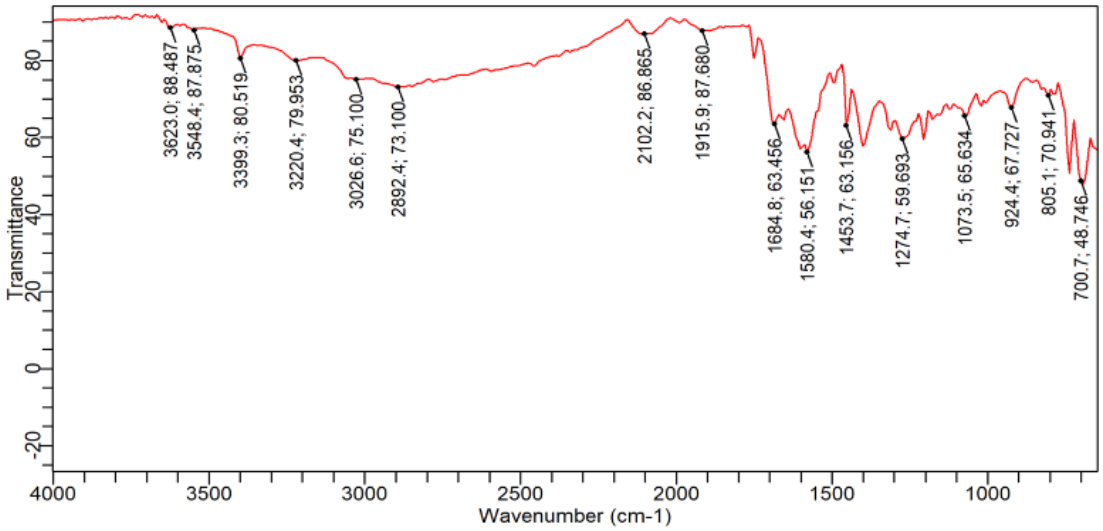


Figure 11: FTIR OF CoL_2

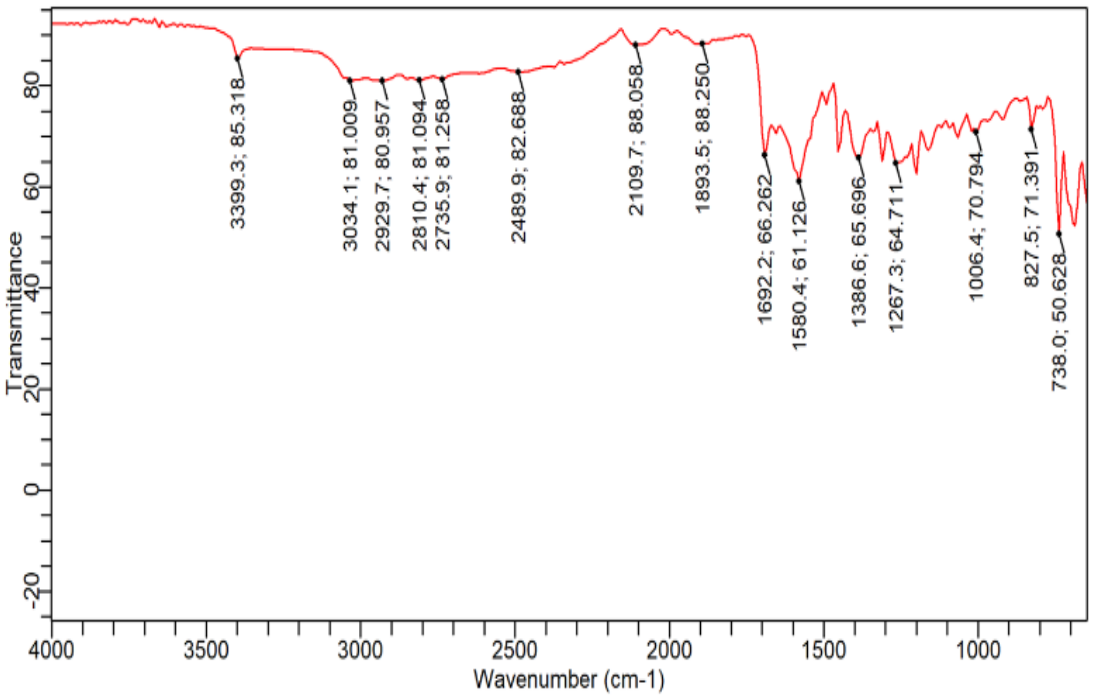


Figure 12: FTIR of Schiff Base (ligand)

The Antihelmintic Profile of the Ligand and Complexes

The antihelmintic study of the ligand BAT and its complexes was carried out *in vitro* using adult tape worm (*Taennia ssp)* with different concentration of 10mg/mL Levamisole (A) as positive control (standard) and water as negative control presented as Figures 13 to 16 . The concentrations of the ligand, complexes, standard and water were considered at 25 %, 50 %, 75 % and 100 % with each corresponding percentage mortality recorded. Each concentration of the ligand, complexes and control was repeated twice for consistency. At 25 % concentration, Levamisole recorded the highest mortality profile of 60% followed by Cu with 30 % while Co complex recorded 20% with water having 0% shown in Figure 13 . At 50 % concentration, the following data were obtained and presented as Figure 14 : ligand 40 %, Cu complex 60 % and Co complex 60 % and Levamisole 90 %. Result obtained at 75 % as figure 15 shows ligand 60%, Cu complex 80 % Co complex 70 % and Levamisole 100 %. Further study at 100 % concentration shown as Figure 16 found that ligand 80 %, Cu 100 %, Co 80 % and Levamisole 100%. It is generally observed that mortality rate of the parasite (*Teannia spp*) is directly proportional to the

concentration of the compounds under study, this agreed with [25]. The graphical representation of these data shown from figure 13-16 from this study has shown that Cu complex have enhanced biological activity than the free ligand as reported by [8], [14] [25]. These findings equally show a constant enhanced biological activity of Cu complex than the Co complex as reported by [25]. It was discovered that Cu complex has the same profile with Levamisole (standard).

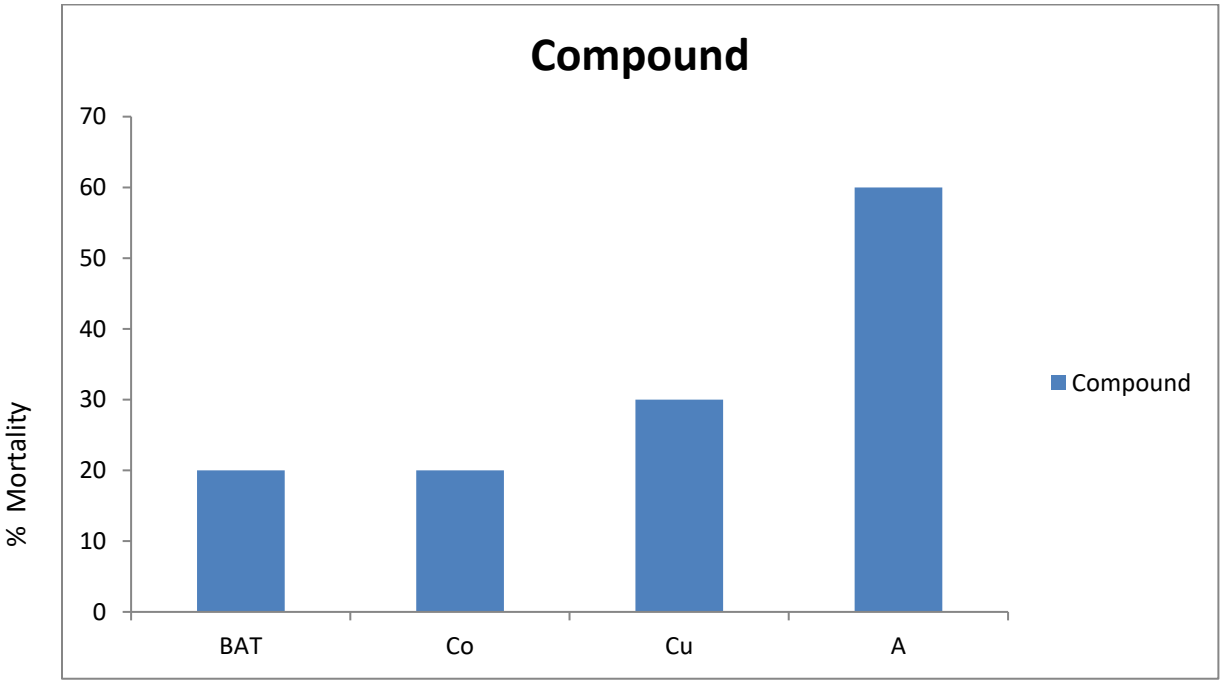


Figure 13: The % Mortality of Tape Worm at 25 % Concentration

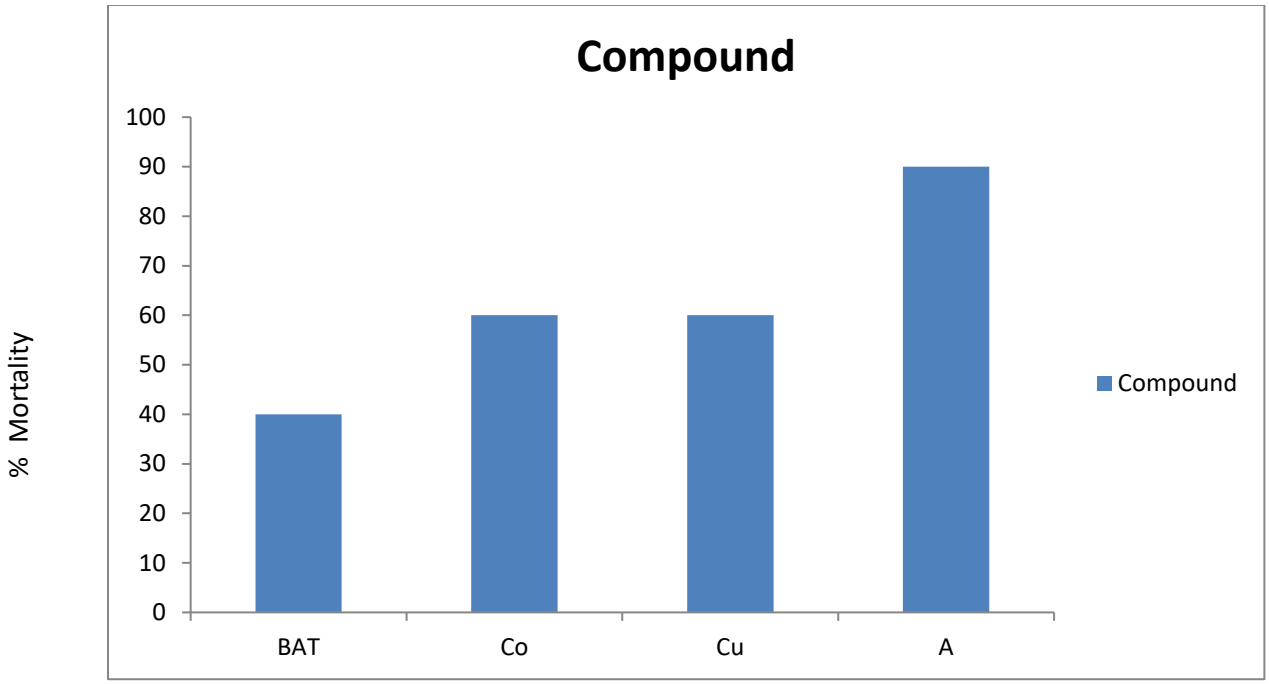


Figure 14: The % Mortality of Tape Worm at 50 % Concentration

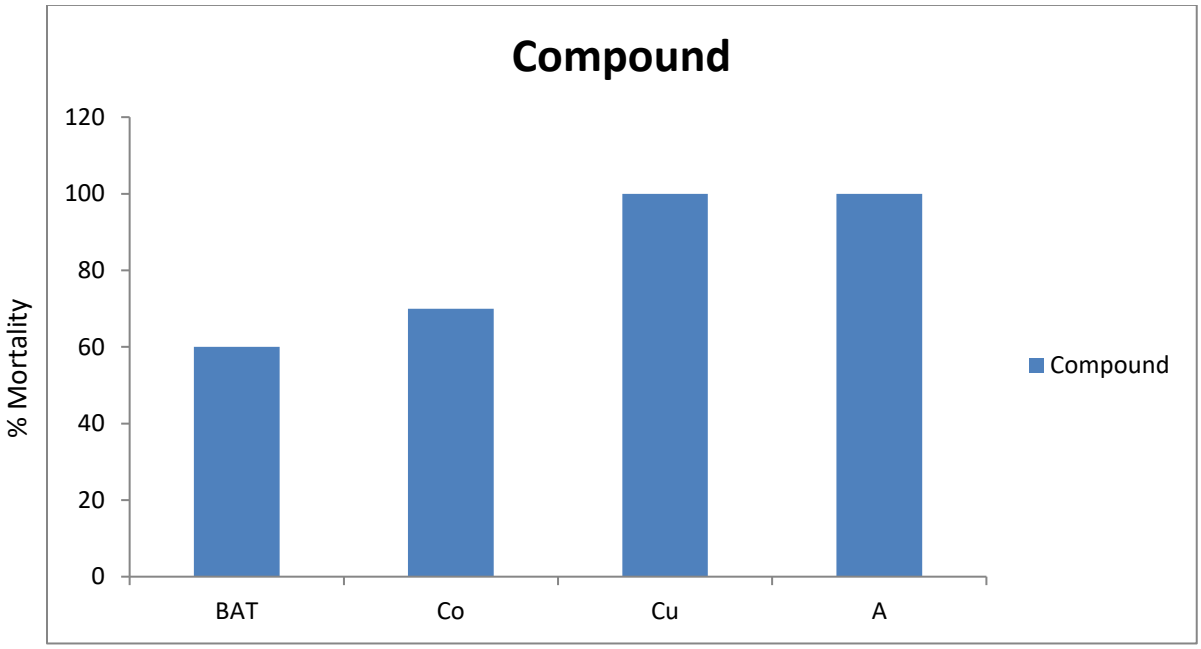


Figure 15: The % Mortality of Tape Worm at 75 % Concentration

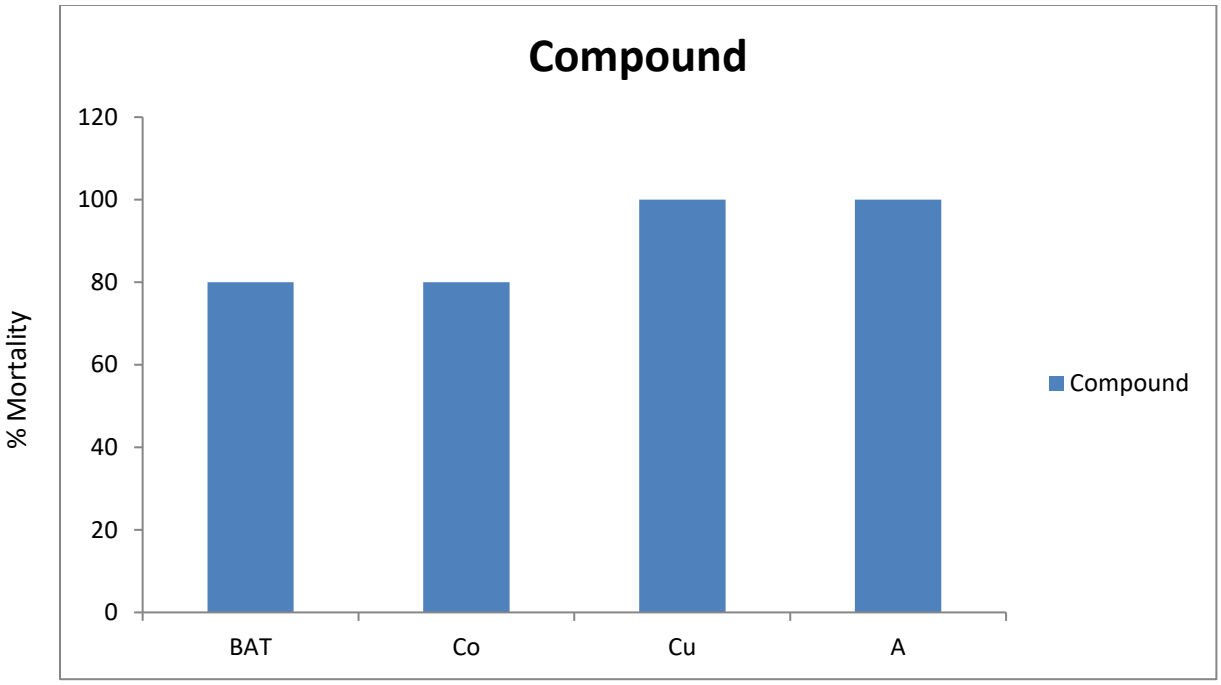


Figure 16: The % Mortality of Tape Worm at 100 % Concentration

Conclusion

In this study, Cu (II) and Co (II) complexes were prepared from (Z)-2-(benzylideneamino)-3- (1H indol-3-yl)propanoic acid Schiff base and characterized using different spectral techniques. The IR spectral data revealed that the Schiff base ligand coordinates through the azomethine nitrogen and carbonyl oxygen atoms in a tetradentate manner. The UV revealed metal to ligand coordination bond. Magnetic and electronic spectra studies revealed octahedral geometry for the complexes. It was observed that the metal chelate in Cu complex exhibit higher antihelmintic property compared to the free ligand. The Cu complex has the same biocidal effect as Levamisole hence; Cu complex can serve as a potential replacement drug for the treatment of tape worm infection.

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