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ORIGINAL STUDY

Preparation and Characterization Using a New Schiff Base Ligand Derived from Benzoyl Isothiocyanate with Their Complexes and Study of Their Biological Activity

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ABSTRACT

Complexes containing Nickel, Copper, and Zinc salts were prepared and studied in this study. Additionally, novel Schiff Base ligands were synthesized by reacting 6-aminopenicillinic acid with benzoyl isothiocyanate to produce L. The synthesized ligand was evaluated by physical measures and spectroscopic methods such as Fourier transform infrared spectroscopy, nuclear magnetic resonance, ultraviolet-visible light, mass spectroscopy, and micro elemental analysis. Metal salts of (Ni(II), Cu(II), and Zn(II)) were combined with novel ligands at a molar ratio of 1:2 [M:L] to prepare the complexes. Spectroscopic, magnetic susceptibility, molar conductivity, atomic absorption, and DTA methods were used to study the complexes that were prepared. The β-lactam ring complexes exhibited coordination with the central metal ion through the N-atoms of the azomethine group and the O-atoms of the carbonyl group, leading to the formation of the resulting octahedral hybrid form, which was denoted as Sp3d2.Experiment on the biological activity of each ligand and its complexes against two bacterial species (S. aureus and P. pseudomonas), and find that all of them are more active than the starting materials.

1. Introduction

An exciting and experimentally challenging field in current chemical sciences, inorganic chemistry relies on coordination chemistry as one of its main pillars.Coordination compounds are currently the focus of the majority of inorganic chemistry research. These compounds have led to the creation of new and exciting products with unique uses in many different industries, such as pharmaceuticals, pesticides, paints, polymers, and photoconductors (Kubra, N.K., et al., [1]). In a coordination complex, a central atom or ion acts as a Lewis acid after forming coordinate bonds with one or more molecules or ions (Ligands) that function as Lewis bases (Soldatović, T., [2]). According to Sinha et al. [3], ligands that have donor atoms directly bonded to the main atom or ion are known as ligands.Chemical substances that include multiple functional groups, such azomethine (C = N) Hugo Schiff, a scientist, was responsible for coining the term “Schiff base” to describe the stable byproduct of condensation between aldehydes or ketones and primary amines. The standard syntactic formula for Schiff bases is RHC = N-R1, and they contain an azomethine group. According to Shah et al. (2020), the groups (-R) can be aromatic, aliphatic, homogeneous, or heterogeneous, among other possible alternatives to Schiff bases. Treatment of diseases in animals and people is facilitated by antibiotics, which kill or restrict the growth of bacteria

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(Vanessa Minden, et al., [4]). The majority of antibiotics used to humans are β-lactam antibiotics (Marco E., et al., [5]). The semisynthetic antibiotics amoxicillin, ampicillin, cefadroxil, and cephalexin, as well as the active pharmaceutical intermediate 6-aminopenicillanic acid (6-APA), are among the most significant medicines in the world. 6-Amino Penicillinic acid is an essential component in the manufacturing of β-Lactam antibiotics, and its demand has been driven up by the rise of antibiotic resistance (Amol M., et al., [6]). One significant pharmacological and therapeutic application of 6-aminopenicillin acid is…The presence of an amino group in the structure of benzoyl isothiocyanate (BI) has made it stand out as a novel comonomer. It is remarkable that amino groups and the benzene ring were bonded in (BI) (Huinan Che., et al., [7]). According to Ali M. S. et al. [8], amino acid derivatives of benzoyl isothiocyanate can be utilized as building blocks for the production of further physiologically active compounds containing a thiourea backbone.

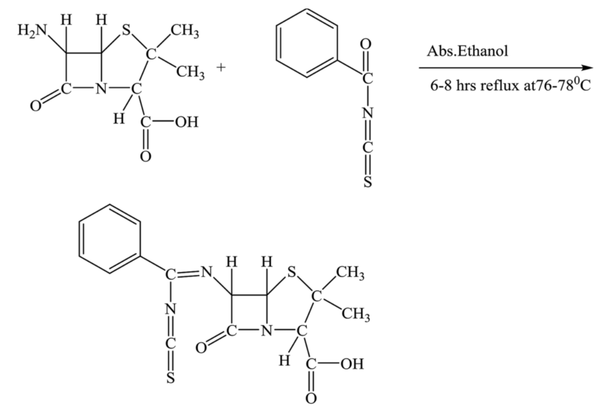
2. Experimental

2.1. Synthesis of ligands (L)

The ligands (Schiff base) were prepared using the general method (Srivastava, A. K., *et al.*, 2021). where (2.162g) from 6-aminopenicilic acid (0.01 mol) was weighed and then dissolved in 50 ml absolute ethanol in around bottom 250 ml. the mixture was heated to 76^{0}C and 4–5 drops of pure pyridine were added, then a volume (1.363 ml) of benzoyl isothiocyanate, was added to the solution to get ligands (L_1), Then the mixture was reflux for period ranging for 6–8 hours at a temperature 76–78^{0}C. A color change from light yellow to dark red in (L) After the reaction was completed, dried the product at room temperature mixture and recrystallized by using Ethanol, Scheme 1 show the preparation reaction for Ligands (L).

2.2. Preparation of complexes with ligand (L)

After weighing the ligand (0.722 g, 0.002 mol) in (L2) and dissolving it in 20 ml of 100% ethanol, the resulting solution was combined with (0.170 g, 0.237 g, and 0.137 g) of $CuCl_2$. $2H_2O$, $NiCl_2$. $6H_2O$, and $ZnCl_2$. (0.001 mol) correspondingly, and let to sit for at least one day. The chemical equations illustrate the production of complexes with ligands when the reactions were finished, the product was allowed to dry at room temperature, filtered, and washed multiple times with distilled water and ethanol (Kuate, M., et al., [9]).



Scheme 1. Preparation of Ligand (Z)-6-((isothiocyanate(phenyl) methylene) amino)-3, 3-dimethyl-7-oxo-4-thia-1-azabicyclo [3.2.0] heptane-2-carboxylic acid (L).

$$2C_{16}H_{15}N_3O_3S_2 + NiCl_2.6H_2O \xrightarrow{EthOH.25^0C} [Ni(C_{16}H_{15}N_3O_3S_2)_2Cl_2]$$
$$2C_{16}H_{15}N_3O_3S_2 + CuCl_2.2H_2O \xrightarrow{EthOH.25^0C} [Cu(C_{16}H_{15}N_3O_3S_2)_2Cl_2]$$
$$2C_{16}H_{15}N_3O_3S_2 + ZnCl_2 \xrightarrow{EthOH.25^0C} [Zn\ (C_{16}H_{15}N_3O_3S_2)_2Cl_2]$$

Table 1. Physical properties of prepared compounds.

Compounds	M.p (°C)	M.wt (g/mol)	Color	Yield%
L	97 ^{0}C	361.43	Black-red	91.5%
]Ni(L)$_2$ Cl$_2$[	120 ^{0}C	852.45	Red	62.8%
]Cu(L)$_2$ Cl$_2$[	102 ^{0}C	857.31	Bronzy	44%
]Zn(L)$_2$Cl$_2$[	112 ^{0}C	859.14	Light Red	78.9%

3. Result and discussion

The physical properties of the prepared compounds, such as melting point, color, molecular Wight and product yield , were calculated, Table 1 shows the results of these calculations.

3.1. FTIR spectrum for ligand and their complexes (L)

The FTIR Spectrum of (6-aminopencilinic acid) exhibited a band 3477.77 cm^{-1} referring to υ(-NH$_2$) group, and the band at 1624.12 cm^{-1} was referred to $\upsilon(-C = O)$ of the β-lactam group (Juan Roberto Anacona *et al.,* 2019), (Bhal, 2015). The FTIR Spectrum for the ligand (L) display of a new band at 1579.75 cm^{-1} assigned to $\upsilon(-C = N)$ for the Schiff base ligand(L), and absence of the band due to $\upsilon(-NH_2)$ suggested the occurrence of the condensation reaction. The band at 1670.41 cm^{-1} was attributed to the stretching vibration $\upsilon(-C = O)$ of β-lactam group (Magdy M. Hemdan., & Amira, A. Alsayed., [10]), (Mohammad Azam et al., 11), (K. Nakamoto, J. W. S., 12]).Supporting Information in Table 2, and the Fig. 1 show the FTIR Spectrum of 6-Aminopenicilic acid, and Schiff base ligands (L).

Table 2. Value of frequencies for the band in FTIR spectrum of prepared ligand (L) and thier complexes.

Compound	υ (OH)	υ (SH)	υ (C = O)	β-lactam υ(C = O)	υ (C = N)	υ (C-N)	υ (C-O)	υ (C = S)	υ (M-N)	υ (M-O)
L	3234.73	2505	1716.70	1670.41	1579.75	1371	1134	1276–1205	–	–
L-Ni	3298.38	2650	1718	1647.26	1577.82	1373	1139	1265–1201	520	414
L-Cu	3242.46	2560	1718.63	1683.91	1570	1373.	1116	1261–1202	599	518
L-Zn	3234.73	2550	1716.70	1647.27	1581.68	1371.43	1139.97	1278–1201	530	420

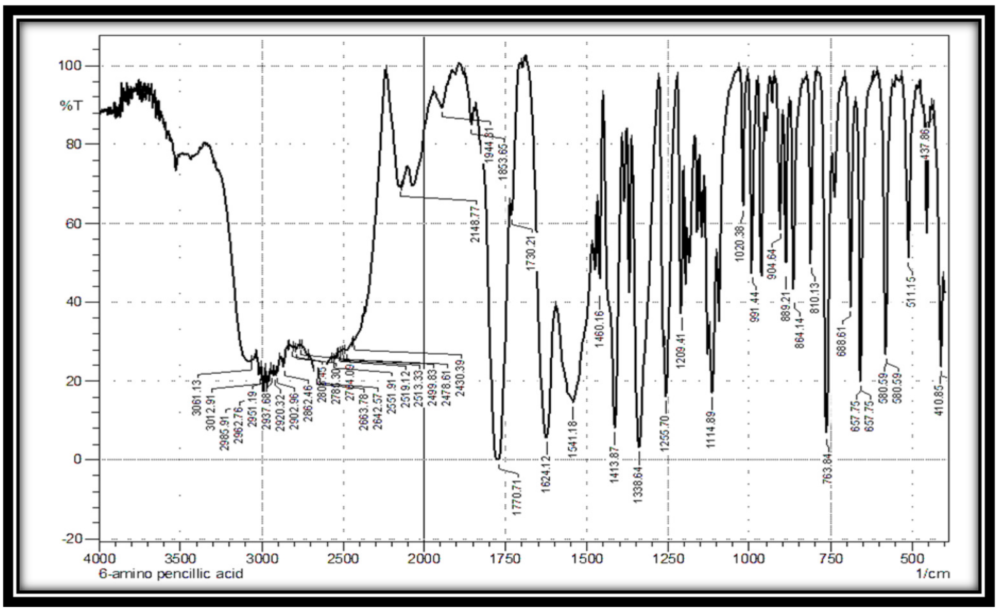


Fig. 1. FTIR spectrum for 6-aminopenicilic acid.

3.2. FTIR Spectrum of Ni(II),Cu(II),Zn(II) Complexes for Ligand (L)

The FTIR spectra of Ni(II), Cu(II), and Zn(II) complexes containing ligands (L) are displayed in Figs. 3 to 5,, respectively. The azomethine group was seen to be involved in the complexation as its frequency was changed to a higher range of 1570–1581.68 cm^{-1} in all complexes, as shown in the figures. Their coordination through the oxygen atom of the υ(C = O) group is shown by the larger shifting of the stretching vibrations at 1647.26–1683.91 cm^{-1} for the υ(C = O) group in the β-lactam ring in the spectra of metal complexes compared to the free ligand. The presence of coordination between the nitrogen atom of the azomethine group and the oxygen atom of the carbonyl group was confirmed by the new weak bands at 599–520 cm^{-1} and 518–420 cm^{-1}, respectively, which were also noted. The frequencies of the bands for the ligand (L) and their complexes are shown in Table 2.

3.3. The ^{1}HNMR spectrum of the ligands (L)

The ^{1}HNMR Spectrum for the ligand (L). Showing Peakes at δ7.3–7.9 ppm (m) ascribed to proton of aromatic ring, and δ8.4–8.5 ppm (d) accredit to the β-lactam ring, The signals at δ 8.6 ppm (s) were observed due to proton of COOH group group (Mohammad Azam., et al., [12]), Fig. 6 shows ^{1}HNMR spectrum for ligand(L).

3.4. The ^{13}CNMR spectrum of the ligands (L)

The ^{13}CNMR Spectrum for (L) Showing peakes at the δ124.39–129.73ppm referred to the aromatic ring, the δ132.59, 133.69 ,136.64 ppm attributed to carbon atom for β-lactam ring, The signals at δ150.05 ppm and δ168.55–168.81 ppm, δ170.25–171.34 ppm (d),190.10 ppm were observed due to azomethine, C = S, C–S and COOH, respectively. (Magdy M. Hemdan & Amira, A. Al-sayed., et al., [13), 2016),(Felix Odame., et al., [11]), (Mohammad Azam et al., [12]), Fig. 7 shows ^{13}CNMR spectrum for ligand (L).

3.5. Micro elemental analysis for ligand

The micro elemental analysis of the prepared ligand (L) was measured. There was a correspondence between the practical ratio value that was the result of experimental work with the theoretically calculated ratios from the relative equations of the elements

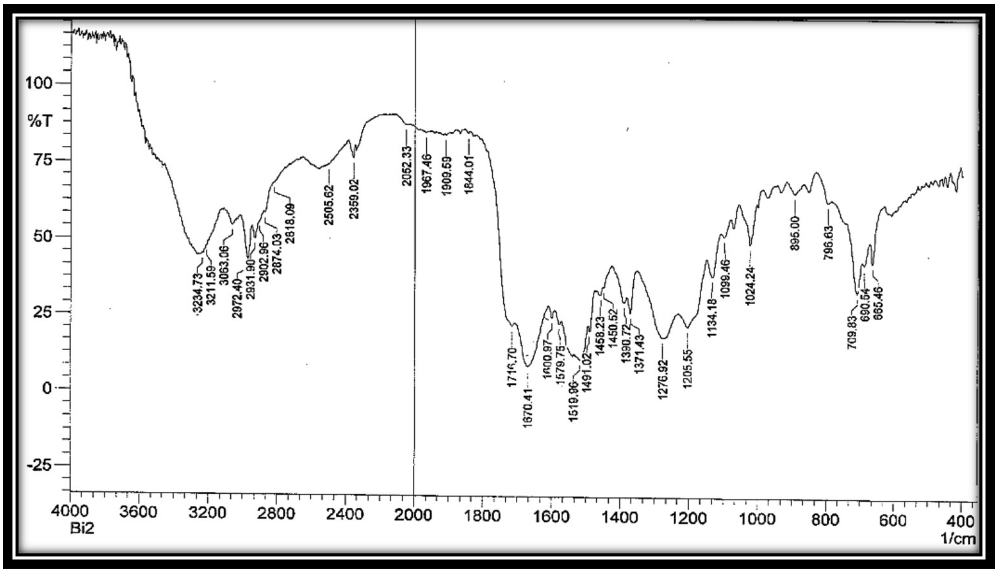


Fig. 2. FTIR spectrum for ligand (L).

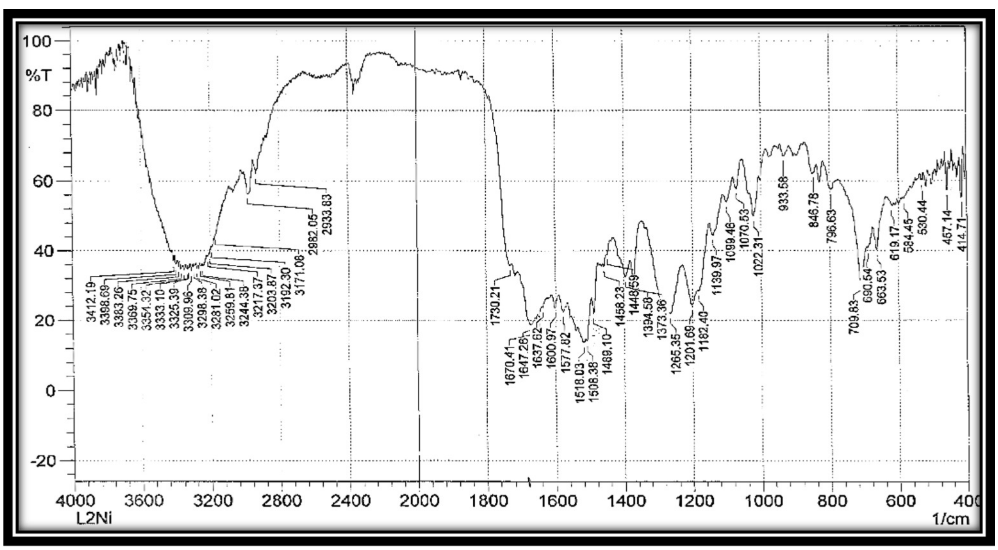


Fig. 3. FTIR spectrum for Ni^{+2} complex with ligand (L).

obtained from this measurement, Table 3 shows the practical and theoretically data of Ligands (L).

3.6. UV-visible spectrum for ligand

The absorption peaks at wave lengths 369 nm, 271 nm, and 238 nm in the UV-Visible spectra of ligand

Table 3. Value for measurements of C.H.N.S technique of ligands (L).

	%C		%H		%N		%S	
Compounds	Exp.	Cal.	Exp.	Cal.	Exp.	Cal.	Exp.	Cal.
L	20.306	17.7	15.22	11.63	7.165	4.15	54.62	53.18

(L) are attributed to the n-π^* transition for the nitrogen azomethene group, the isothiocyanat group, and unsaturated bonding, respectively (Issa, Y. M., et al., [14]). Fig. 8 shows the UV-Visible spectrum of ligand (L).

It was discovered that in Ni(II) and Cu(II) complexes, the azomethine group shifted to a lower wavelength at 366.2 nm and 214.5 nm, respectively, (Blue shift), as a result of the ligand field transitions (L.F.), when the ligand spectra were compared with those of the complexes. A new band at 231.5 nm is observed in the Cu(II) complex as a result of the 2Eg $\rightarrow$ 2T2g transition, while a band at 369 nm is lost in the spectrum of the free ligand.

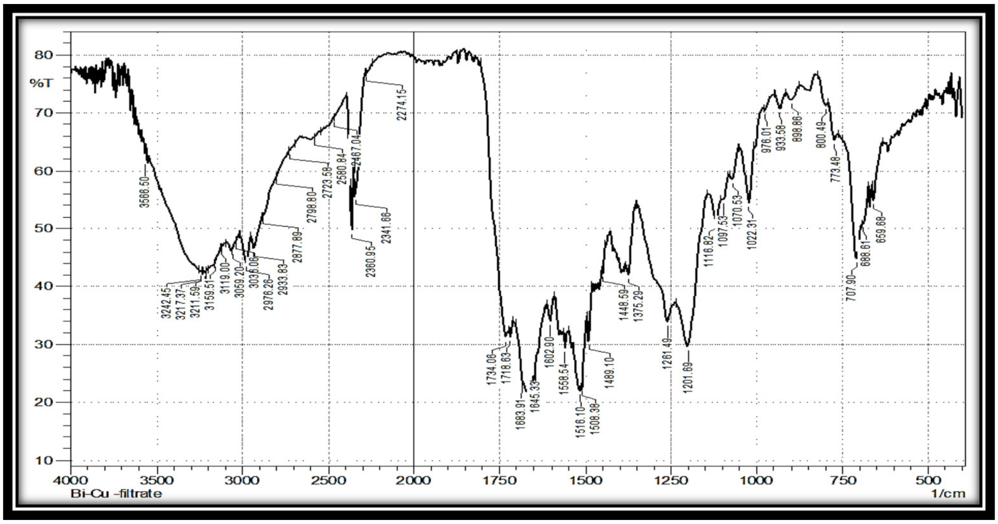


Fig. 4. FTIR spectrum for Cu^{+2} complex for ligand (L_2).

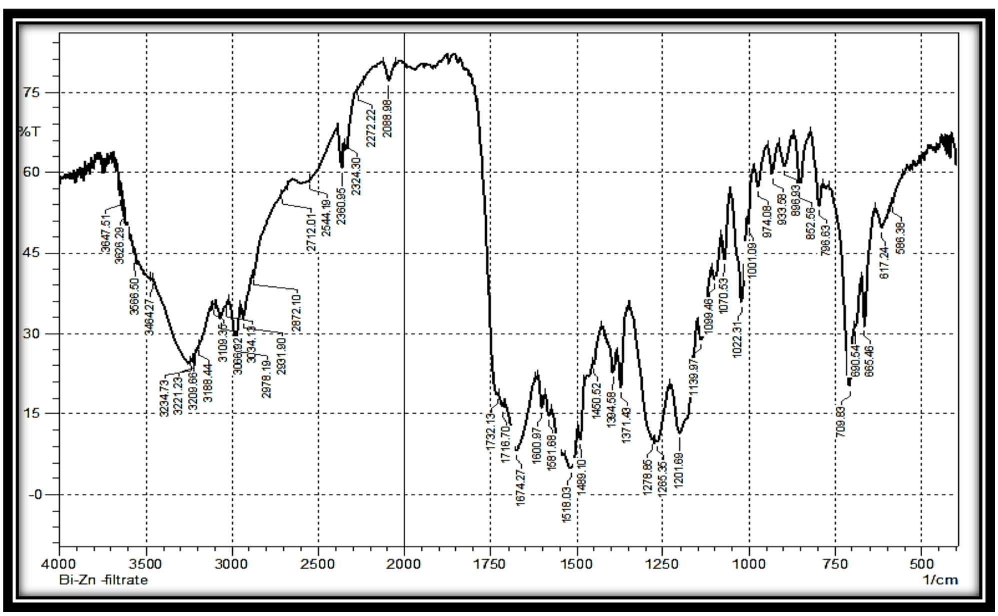


Fig. 5. FTIR spectrum for Zn^{+2} complex with ligand (L).

The azomethine group shifted to a higher wavelength at 369.2 nm in the Zn(II) complex spectra, while a new band at 210.2 nm was observed as a result of charge transfer and ligand field transitions, respectively (Ibtisam M Ali., et al., [20]). The UV-Visible spectra of the Ni(II), Cu(II), and Zn(II) complexes with ligand (L) are shown in Figs. 9 to 11 respectively, and the data is listed in Table 4.

Table 4. Electronic transition of the ligand(L) and its complexes.

Compounds	λ_{max}	$\upsilon_{cm}{}^{-41}$	Assignment	Shifted
L	369 nm	2710 cm^{-}	n-π^*	
	271 nm	3690 cm^{-}	π- π^*	
	238 nm	4201.68 cm^{-}	π- π^*	
$[Ni(L)_2Cl_2]$	366.2 nm	2730.74 cm^{-}	L.F	Blue shift
$[Cu(L)_2Cl_2]$	231.5 nm	4319.65cm^{-}	^2Eg-$^2T_{2g}$	Blue shift
	214.5 nm	4662 cm^{-}	L.F	
$[Zn(L)_2Cl_2]$	369.2 nm	2708.55 cm^{-}	L.F	Red shift
	271 nm	3690 cm^{-}	L.F	
	238.3 nm	4196.4 cm^{-}	C.T	
	210.2 nm	4757.4cm^{-}	C.T	

3.7. Mass spectrum for ligand (L)

The mass spectrum of the ligands (L)shows the appearance of a peak at (360 M/Z^{+}) which is attributed

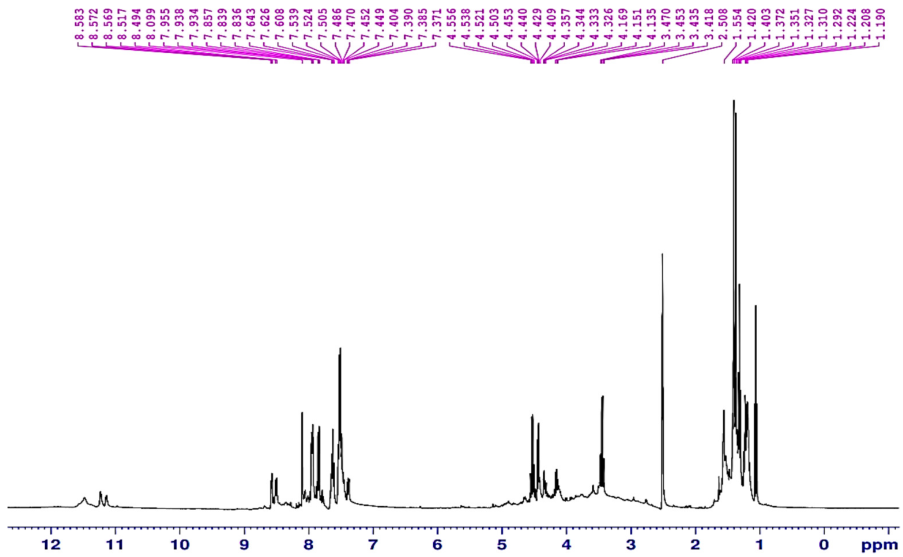


Fig. 6. [1]HNMR spectrum for ligand (L).

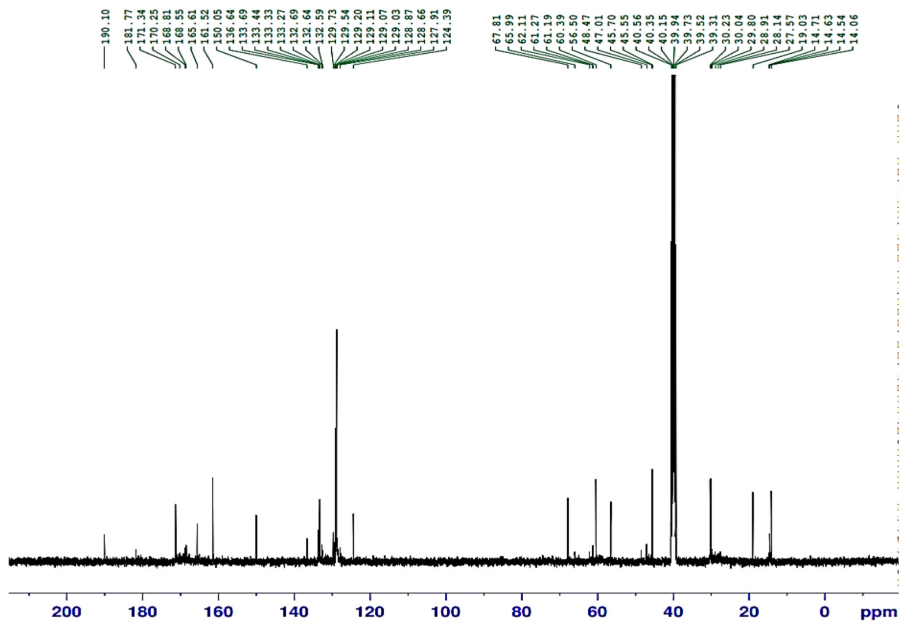


Fig. 7. [13]CNMR spectrum for ligand (L).

to the parent molecular ion, which corresponds to the proposed molecular formula of the ligands(L) These spectrum recorded other peaks, most of which agree with what was mentioned in the prevue's studies (Rasha Hasan Jasim., et al., [16]). the diagrams below Scheme 2 and shows the proposed fragmentations path for two ligand and Fig. 12 shows the mass spectrum of the ligand (L).

3.8. Atomic absorption spectroscopy

The results have shown convergence between practical and theoretical values, this indicates the correctness of the proposed formulas, as well as the correctness of the mixing ratios between metals and ligand. Table 5 shows theoretical and Practical values for the ratio of metal to the ligand and the suggested formula of the prepared complexes.

3.9. Thermogravimetric analysis (TGA)

Thermogravimetric analysis (TGA) is a helpful method for figuring out the thermal stability and breakdown mechanism of complexes.

The TG and differential thermal analysis led to the proposal of the Ni(II) complex structural formula.

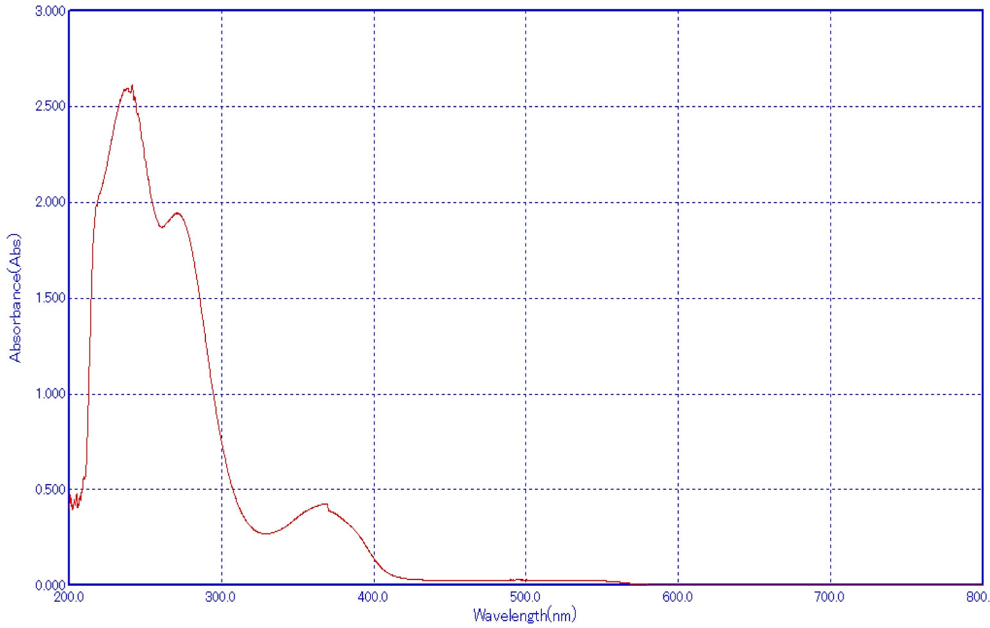


Fig. 8. UV-visible spectrum for ligand (L).

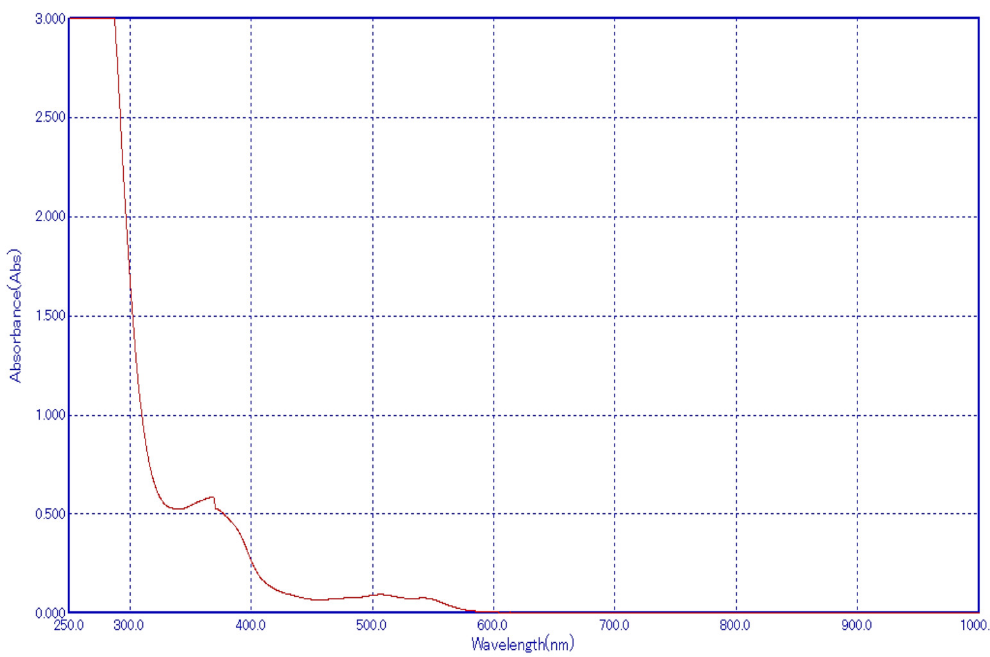


Fig. 9. UV-visible spectrum for Ni^{+2} complex with ligand (L).

There was a single main breakdown stage for the Nickel (II) complex, according to the TGA curve. Within the temperature range of 191.30–216.850C, the complexes mass loss occurred, with a weight loss of 22.643% and a differential thermo gravimetric maximum (DTGmax) of 202.940C. As shown in (Fig. 13), the TGA curve for Ni(II) ion complexes with (L) ligand is devoid of water molecules both within and outside the coordination sphere for nickel complexes. Our measurement proves it.

The TGA curve showed that there was a single primary breakdown stage for the copper (II) complexes. Within the temperature range of 206.11–213.17C°, the greatest mass loss occurred, resulting in a weight loss of 50.774% and a DTGmax of 180.04C0. For copper complexes, this measurement proves that no

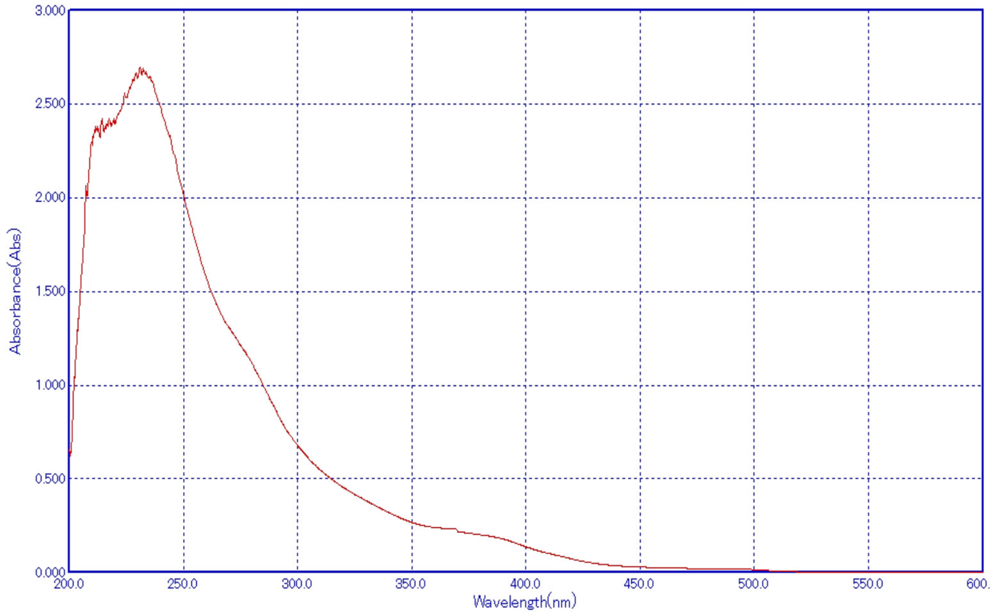


Fig. 10. UV-visible spectrum for Cu^{+2} complex with ligand (L).

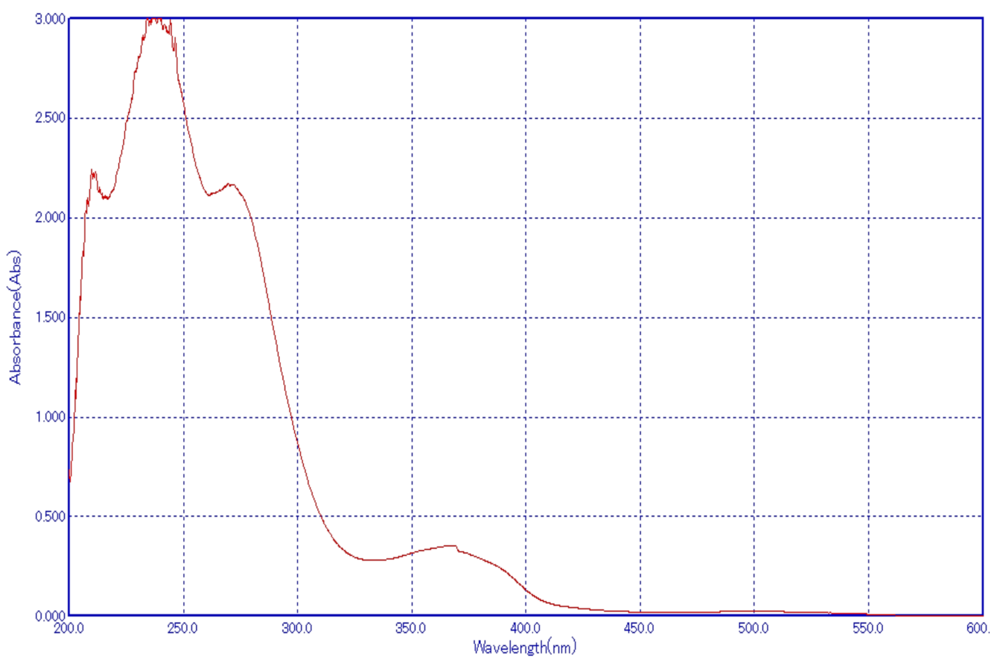


Fig. 11. UV-visible spectrum for Zn^{+2} complex with ligand (L).

Table 5. Theoretical and practical values for the ratio of metal ions in the complexes, and suggested formula of the prepared complexes.

Complexes	%Metal		Suggested formula
	Exp.	Cal.	
L-Ni	6.3	6.9	]Ni $(L)_2Cl_2$[
L-Cu	11.4	7.4	]Cu $(L)_2Cl_2$[
L-Zn	8.8	7.6	]Zn$(L)_2Cl_2$[

water molecules are present either inside or outside the coordination sphere. This is illustrated by the TGA curve for the Cu (II) complex of ligand (L) in Fig. 15.

The Zinc(II) complex decomposed in one primary breakdown stage, according to the TGA curve. With a DTGmax of 189.85 °C and a temperature range of 177.07-217.26 °C, the complexes' mass loss in the stage was 38.845%. The TGA curve for Zn (II) complexes of ligand(L) is show in Fig. 16 and

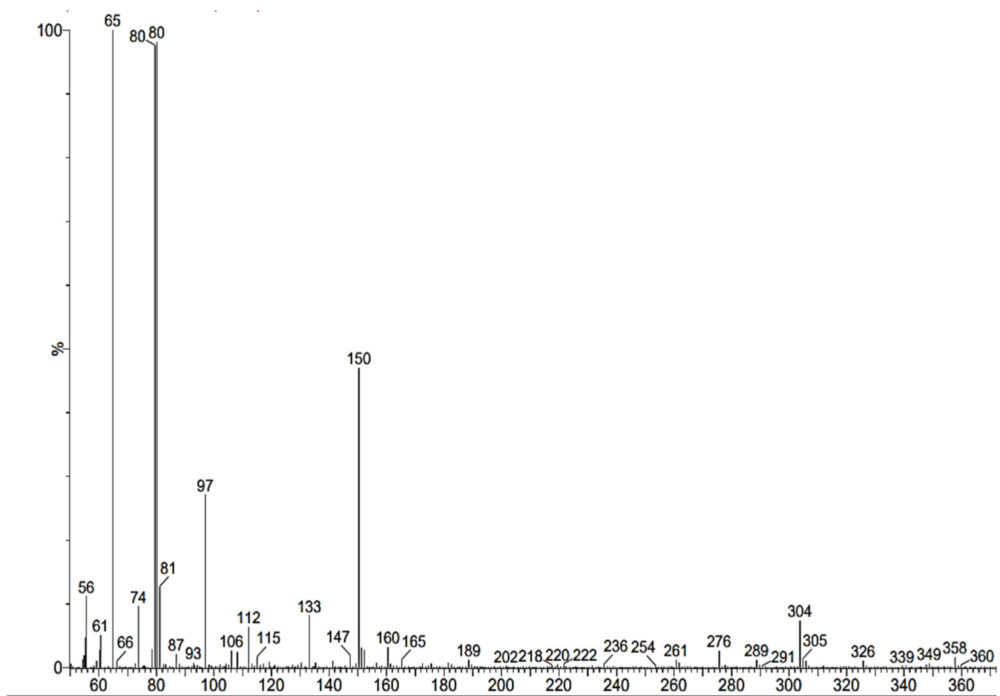
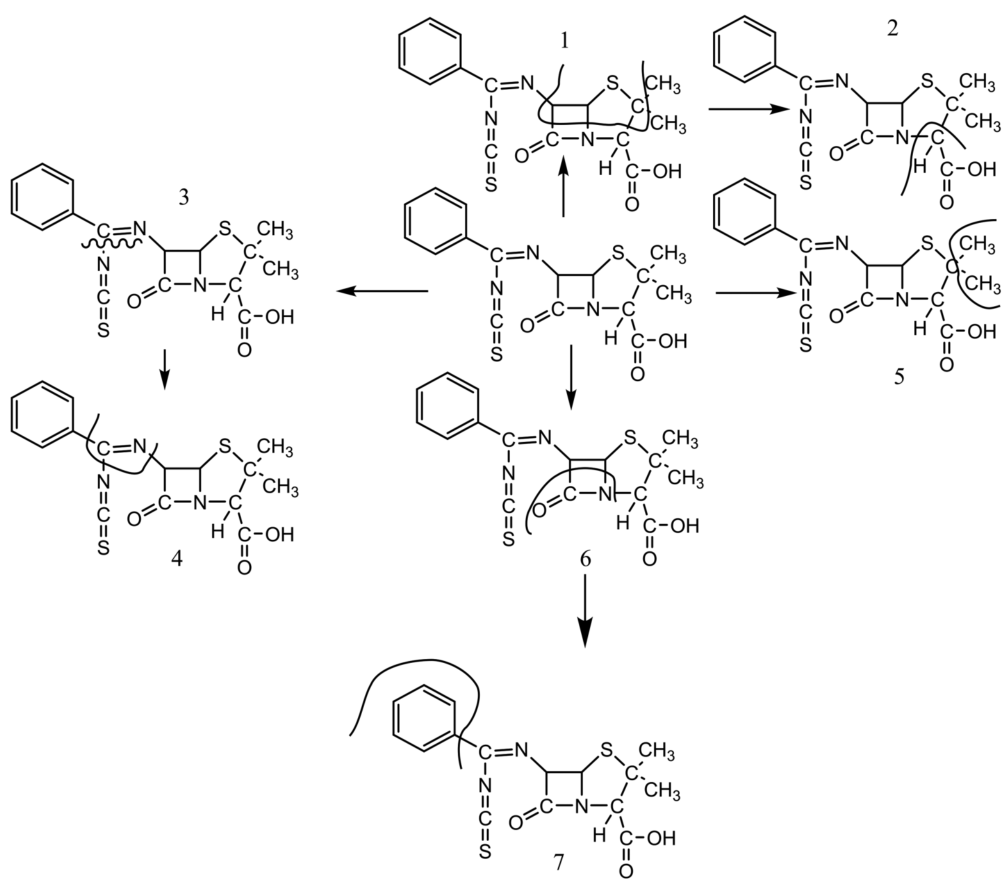


Fig. 12. Mass spectrum of ligand (L).



Scheme 2. Fragmentation of the ligand(L).

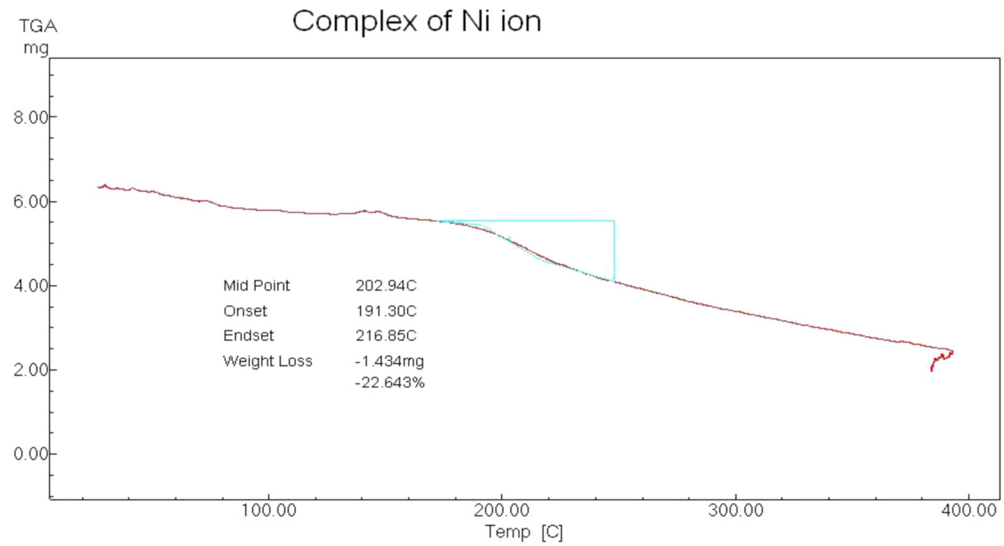


Fig. 13. Thermogravimetric analysis curve for Ni^{+2} complexe with Ligand (L_2).

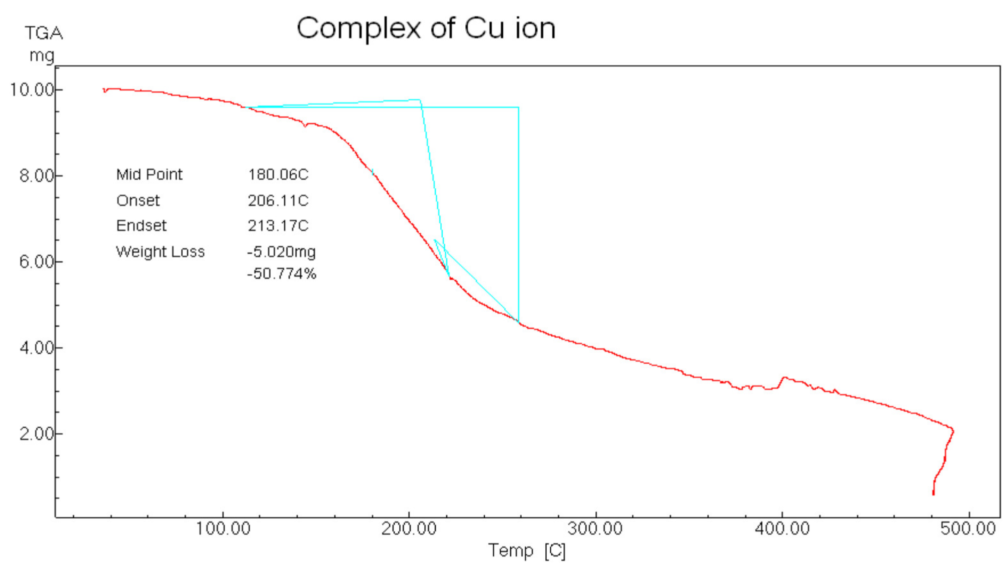


Fig. 14. Thermogravimetric analysis curve for Cu^{+2} complexes.

this measurement demonstrates that there are water molecules not present both inside or outside the coordination sphere for zinc complexes.

3.10. Magnetic susceptibility and molar conductance for coordination complexes

The complexes $[Ni(L)_2\ Cl_2]$, $[Cu(L)_2Cl_2]$, and $[Zn(L)_2Cl_2]$ were determined to have magnetic susceptibility values of (0.17, 2.327, and 0 M.B.) when compared to previous research, which indicated that the octahedral shape was most common when these complexes were formed. According to N. Ghufran Kareem, M.H.S. [17], the octahedral shape can be achieved by coordinating metal ions with the oxygen atoms of the carbonyl group and the nitrogen atom of the azomethine group in a bidentate ligand (L), as shown in Table 6.

The octahedral shape is achieved through the ligand's (L) bidentate ligand interaction with Sp3d2 hybridization, as illustrated in Fig. 17.

The antimicrobial effects of recently produced chemicals. This approach states that the quantity of microbes grows as microbial cell growth rates rise (Taghreed H. Al-Noor., et al., [18]). (Sawant, Amol M., et al. [6]). The biological activity of four different types of bacteria (Staphylococcus aurous and Pseudomonas) was tested with the ligand complexes

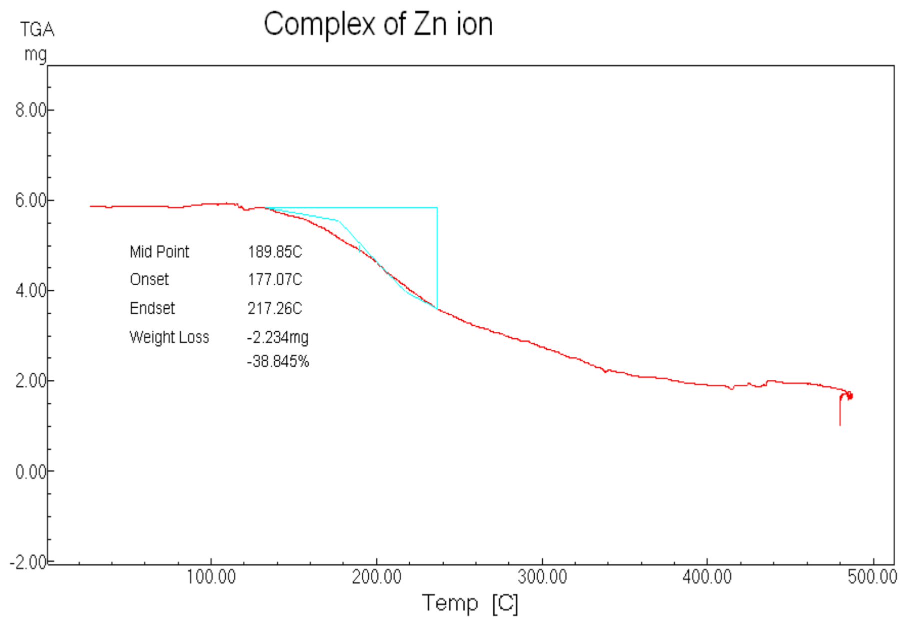


Fig. 15. Thermogravimetric analysis curve for Zn^{+2} complexe with ligand (L_2).

Table 6. Magnetic susceptibility and conductivity at the molar level for coordination complexes.

Complexes	μ eff (B.M) Cal.	Exp.	Molar conductance (Ω^{-1} cm^2 mol^{-1})	Shape
$[Ni(L)_2Cl_2]$	2.82	0.17	10	Oct
$[Cu(L)_2Cl_2]$	1.73	2.327	2	Oct
$[Zn(L)_2Cl_2]$	0	0	0	Oct

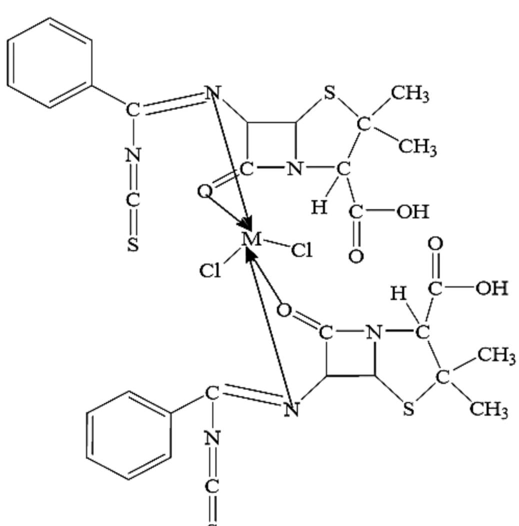


Fig. 16. Suggested shape of metal ion complexes with ligand (L), M = Ni(II), Cu(II), and Zn(II).

with Ni(II), Cu(II), and Zn(II) at a concentration of 1×10^{-3} M. The results were compared to the inhibitory effect of the original ligand (L) on the same

Table 7. Minimum inhibitory concentration (MIC) in (g/mL) of the ligands and their metal complexes.

Compounds	*pseudomonas*	*Staphylococcus*
2-L	15	0
5-$[Ni(L)_2\ Cl_2]$	15	12
3-$[Cu(L)_2Cl_2]$	13	15
4-$[Zn(L)_2Cl_2]$	16	12

bacteria. When the ligand and its complexes were applied to the plantation of pseudomonas (Fig. 13) and Staphylococcus aureus (Fig. 14), the inhibition ability of the four complexes was higher than that of the original ligand, which was studied at a concentration of 1×10^{-3}. (in the 5th table). Among the three complexes and the original ligand, the zinc ligand (L2) exhibited the strongest inhibitory effect (Catia Longhi et al., [19]). In contrast, the copper complex exhibited slightly stronger inhibitory effects at a concentration of 1×10^{-3} M compared to the nickel complex at the same concentration (Ryldene Marques Duarte da Cruz et al., [15]). (1) 10^{-3} millimolar Table 6. Measured in milligrams per milliliter, the ligand and metal complex minimum inhibitory concentration (MIC).

4. Conclusion

According to Catia Longhi et al. [19], 6-aminopenicillanic acid (6-APA), a key component of penicillins and an active pharmacological intermediate, has the potential to be transformed into semisynthetic antibiotics. A Schiff base reaction was used to synthesis the ligands (L2) from

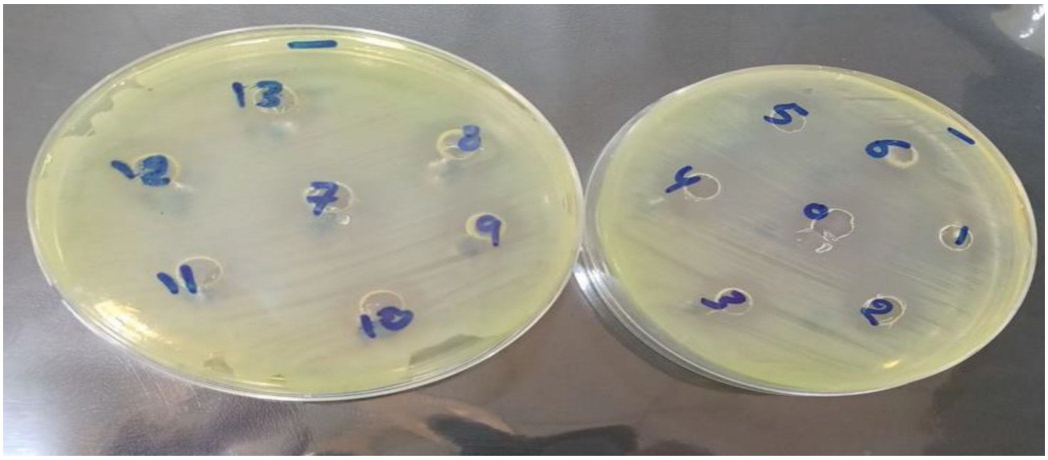


Fig. 17. Ligands (L_1, L_2, L_3) and its complexes were applied on the plantation of *Staphylococcusaureus*.

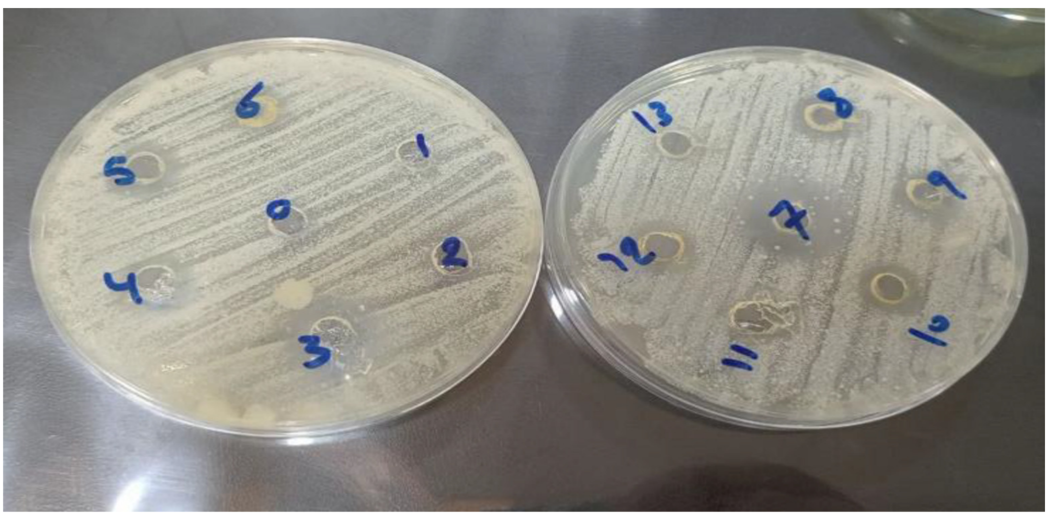


Fig. 18. Ligands (L_1, L_2, L_3) and its complexes were applied on the plantation of *pseudomonas*.

6-aminopenicillin acid and benzoyl isothiocyanat, resulting in ((Z)-6-((isothiocyanato(phenyl) methylene)amino). Ligand consisting of (3, 3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0] heptane-2-carboxylic acid) and In a molar ratio of [1:2] for the Schiff base produced from this ligand, three novel metal(II) complexes were formed. According to FTIR measurements, the Schiff base acted as a monoanionic NO-chelating agent with azomethine-N and carbonyl(β-lactam)-O atoms, forming a bidentate configuration. The molar conductance of the metal complexes ranged from 0 to 10 Ω−1 cm2 mol−1, indicating that they were not electrolytes. The octahedral geometry was revealed by the electronic spectra of the Ni(II), Cu(II), and Zn(II) complexes. To determine whether the Schiff base and its metal complexes inhibited the growth of staphylococcus and pseudomonas, researchers conducted tests.

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