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Spectroscopic and Quantum Chemical Studies of Some Novel Mixed-ligand Complexes of Fe(II) and Cu(II) and Investigation of their Antimicrobial actvities

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ABSTRACT

This study reports the synthesis, spectral characterization, quantum chemical studies, and biological activities of homo- and heteroleptic Fe(II) and Cu(II) complexes derived from 2-(4-bromobenzylidene)-aminophenol (L_1). The ligand was prepared via a condensation reaction and characterized using FT-IR, UV-Visible spectroscopy, CHNS analysis, ^1H NMR, and ^{13}C NMR. The complexes were synthesized in a 1:1 metal-to-ligand (M:L) ratio and characterized by melting point, UV-Vis, FT-IR, molar conductivity, and magnetic susceptibility measurements. FT-IR data confirmed bidentate coordination through oxygen and amide atoms, forming neutral complexes. Magnetic moments and electronic spectra support octahedral geometries for both Fe(II) and Cu(II) complexes. Molar conductance measurements indicated nonelectrolyte behavior in methanol. Key parameters from DFT (Density Functional Theory) calculations at the B3LYP/6–311++G(d,p) level are also reported. The Gauss View molecule imaging program was utilized to visualize the input data files as well as determine the HOMO–LUMO energies. The complexes exhibited notable antibacterial activity against both *Salmonella* and *Staphylococcus*. However, complexes C_3 and C_4 showed reduced activity against both bacteria, indicating that the homoleptic complexes of this ligand possess greater antibacterial efficacy. The ligand (L_1) itself showed negligible activity against either bacterium. These results highlight the significant role of metal coordination in influencing biological activity.

Keywords: Spectroscopic, Quantum chemical, Coordination, *Staphylococcus*, Mono-nuclear, Complexes, Octahedral geometry.

INTRODUCTION

Schiff bases represent an important class of organic compounds formed via condensation reactions between aldehydes or ketones and primary amines, first reported by Hugo Schiff[1-5]. These compounds can coordinate with metals through nitrogen and oxygen atoms to form stable complexes[6-7]. When a single type of ligand reacts with metal salts, homoleptic complexes are formed, whereas reactions involving two different ligands with metal salts yield heteroleptic complexes[8], which have important applications as antitumor, antiviral, antifungal, and antibacterial compounds[7,9-10].

Furthermore, mixed ligand complexes of Cu(II), Ni(II), and Zn(II) were synthesized utilizing thiophene-2-carbonyl-isonicotinohy-drazone as the principal ligand, which is prepared from the reaction of isoniazid (INH) with Thiophene-2-carbaldehydein which is mixed with 1,10-phenanthroline hyFor instance, Kudrat-E-Zahan reported the synthesis of Ni(II) and Cu(II) mixed-ligand complexes using 2,2'-bipyridine and a Schiff base ligand derived from isoniazid and anisaldehyde. Similarly, Abd El-Halim[11] prepared mixed-ligand complexes of Mn(II), Co(II), Cu(II), Fe(III), Ni(II), Zn(II), and Cd(II) using a Schiff base synthesized from quinoline-2-carboxaldehyde and 2-aminophenol (1:1 ratio), subsequently combined with 1,10-phenanthroline (1:1 molar ratio)[12]. Shane M. Wilkinson described the preparation of Fe(III) and Cu(II) homoleptic complexes using a Schiff base derived from benzhydrazide and salicylaldehyde. Production of Cu(II) mixed ligand complexes utilizing a tridentate Schiff base ligand and 2,2′-bipyridine, which are used as an antibacterial compound, Zeinab Albobaledi[13]. synthesized mixed-ligand complexes of Cu(II), Co(II), Ni(II), and Zn(II) using a Schiff base prepared from curcumin and 2-aminobenzothiazole, which was then combined with 1,8-diaminonaphthalene. Abdel-Rahman[14]. Formation of mixed ligand complexes regarding Cu(II), Co(II), Ni(II), as well as Zn(II) through the reaction of a Schiff base ligand synthesized via curcumin with 2-amino benzothiazole and this ligand mixed with 1,8-diaminonaphthalene for preparation of mixed ligand complexes Chandrasekar[15]. Preparation of complexes of Mn(II), Fe(III) and Cu(III) its complexes have applications in antibacterial and anticancer, molecular docking, and ADMET studies Abdel-Rahman[16].

Drate is mixed Ashrafuzzaman, M.D[17]. Schiff bases and their metal complexes serve dual roles as industrially valuable agents and biologically active compounds, notably as antifungals[18] and demonstrate a wide array of biological activities, including antifungal properties[10]. They also function as highly effective catalysts across various reactions, though their performance can be compromised by moisture[19].

Recent research shows that coordinating metals to Schiff-base ligands strongly influences both their biological activity and chemical properties. Because Schiff bases can bind metals at multiple sites, their cooperative behavior is being intensely studied. Consequently, designing new Schiff-base ligands and their metal complexes as potential drugs is now a major focus in medicinal chemistry.[20] In this article, we aim to compare homoleptic and heteroleptic complexes prepared from a Schiff base ligand and evaluate their antibacterial activities.

EXPERIMENTAL METHODS

Instruments and Measurements

FT-IR spectra were recorded using a Shimadzu ATR-FTIR spectrometer equipped with a single-bounce diamond ATR accessory at a resolution of 4 cm^{-1}. UV-Vis spectra were obtained using a double-beam spectrophotometer (model 6705 UV, JENWAY, Japan) connected to UV Probe software and interfaced with a computer. All measurements were performed using 10 mm quartz cuvettes with a spectral bandwidth of 1 nm. Elemental analyses (CHNO) were carried out using a Thermo Fisher Eager 300 analyzer. Melting points were determined using an AELAB DMP-800 colorful touchscreen melting point apparatus with open-ended glass capillaries. Magnetic susceptibility measurements were performed at 296.15 K using a Sherwood Scientific magnetic susceptibility balance.

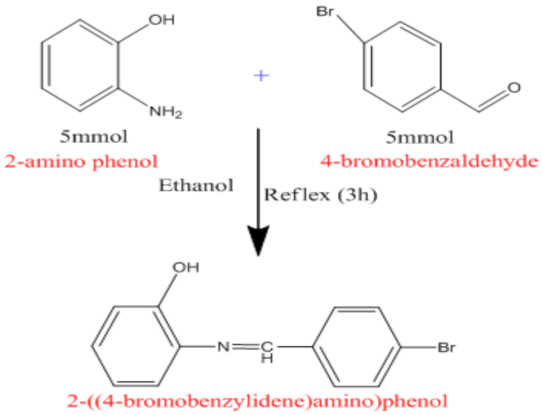
Molar conductivities of 10^{-3} M solutions of the metal complexes were measured at 296.15 K using a TRANASINSTRUMENTSBC3020ProfessionalBenchtop Conductivity Meter in ethanol containing a few drops of DMF. ^{1}H-NMR and ^{13}C-NMR spectra were recorded at room temperature using a Bruker 300 MHz NMR spectrometer in DMSO-d_6, with tetramethylsilane (TMS) as the internal reference.

Material

All chemicals (2-aminophenol, 4-bromobenzaldehyde, CuCl′, FeCl′, ethanol, DMF) were obtained from Shanghai Macklin and used as received without further purification.

Synthesis of Ligand

The Schiff base ligand (L_1), 2-((4-bromobenzylidene)amino)phenol, was prepared by refluxing equimolar solutions of 4-bromobenzaldehyde (5 mmol, 0.925 g) and 2-aminophenol (5 mmol, 0.54 g) in ethanol (8 mL each) at 70°C for 3 hours. Cooling afforded yellow crystals that were filtered, washed with cold ethanol, and recrystallized from ethanol to give pure L_1 in 70% yield, melting piont. 133–137°C. The compound was fully characterized by UV-Vis, $^{1}H/^{13}C$NMR, FT-IR, and elemental analysis[21].



Scheme 1. Synthesis of Schiff base ligand (2-((4-Bromobenzylidene)amino)phenol (L_1)

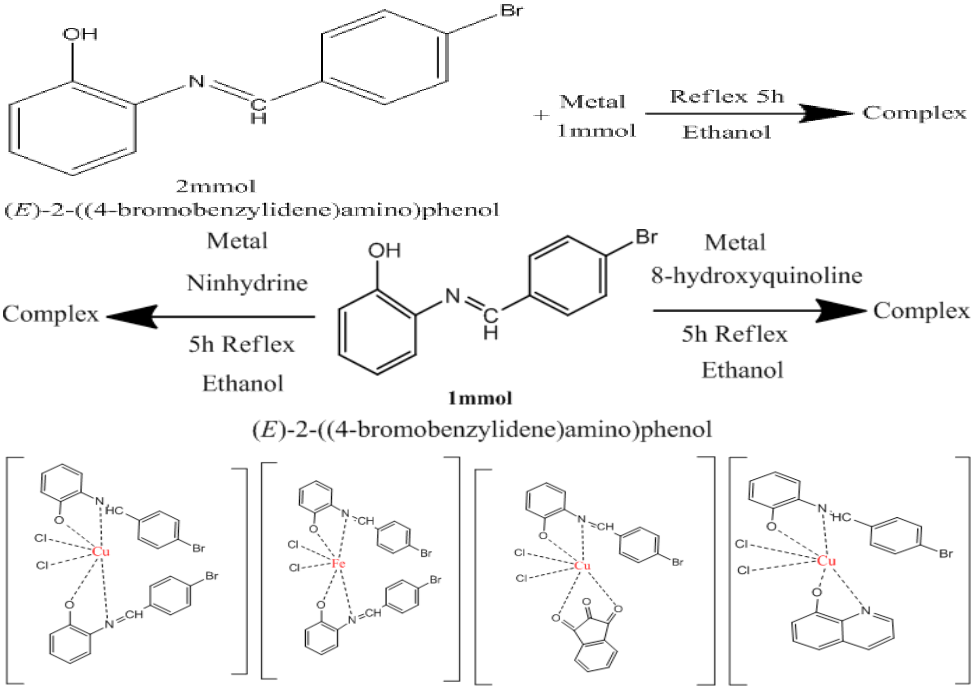
Homoleptic Complexes (C_1 and C_2)

The homoleptic complexes of Cu(II) (C_1) and Fe(III) (C_2) were prepared by reacting the respective metal salts (CuCl′ and FeCl′) with the Schiff base ligand 2-((4-bromobenzylidene)-amino)phenol (L_1). In a typical synthesis, 1 mmol of the metal salt was dissolved in ethanol and mixed with 2 mmol (0.55 g) of L_1 dissolved in ethanol. The reaction mixture was refluxed at 50°C for 5 hours[22]. The resulting complexes were filtered, washed with cold ethanol, and recrystallized from ethanol to ensure purity.

Synthesis Heteroleptic Complexes (C_3 and C_4)

Complex C_3: The heteroleptic Cu(II) complex (C_3) was synthesized by reacting CuCl′ (1 mmol) dissolved in ethanol with a mixture of L_1 (1 mmol, 0.275 g) and 8-hydroxyquinoline (L_2, 1 mmol, 0.145 g) dissolved in ethanol. The reaction mixture was refluxed at 50°C for 5 h[23], Complex C_4: The heteroleptic Cu(II) complex (C_4) was prepared by reacting CuCl′ (1 mmol) dissolved in ethanol with a mixture of L_1 (1 mmol, 0.275 g) and ninhydrin (L_3, 1 mmol, 0.178 g) dissolved in ethanol. The reaction mixture was refluxed at 50°C for 5 hours.

All complexes were purified by recrystallization from ethanol. The synthetic route for all complexes is illustrated in Scheme 2. The yield, color, and melting points of the complexes are summarized in Table 3.



Scheme 2. Synthesis of mixed-ligand complexes of Cu(II) and Fe(II)

RESULTS AND DISCUSSION

Ultraviolet-visible spectra

The UV-Vis spectra of the ligand (L_1) and its metal complexes were recorded at ambient temperature in ethanol solution. The spectrum of the free ligand (L_1) exhibits three distinct absorption peaks: at 207 nm (assigned to n→π* transitions), 272 nm and 354 nm (both attributed to π→π* transitions)[24] as shown in Fig. 1. Upon complexation, the spectra of the metal complexes (C_1-C_4) show additional absorption bands in the visible region: C_1: 450 and 470 nm, C_2: 440 nm, C_3: 400 nm, C_4: 400 and 480 nm.

These new bands correspond to metal-to-ligand charge transfer (MLCT) transitions, confirming coordination between the metal ions and the ligand. Furthermore, weak d-d transition bands are observed at approximately 500 nm (for C_1, C_3, and C_4) and 510 nm for (C_2), which are characteristic of transition metal complexes.

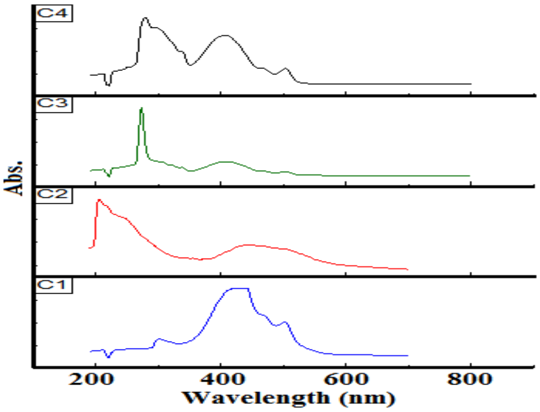


Fig. 1. UV-Visible Spectrum of Schiff base ligand (L_1)

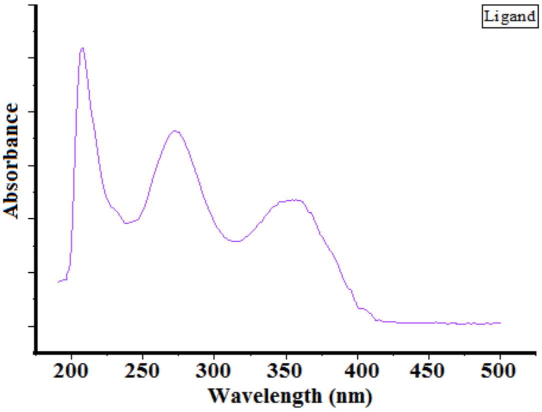


Fig. 2. UV-Visible Spectra of complexes (C_1- C_4)

Table 1: Electronic spectral assignment of ligand (L1) and Complexes (C1-C4)

Compounds	Intra-ligandtransition $'_{max}$ nm	MLCT $'_{max}$ nm	d-d transition $'_{max}$ nm	Assignment
L1	272,354, 207	-	-	-
C1	300	450,470	500	$d_{xy} \longrightarrow dX^2y^2$
C2	200,220,250	440	510	$d_{xy} \longrightarrow dX^2y^2$
C3	270, 340	400	500	$d_{xy} \longrightarrow dX^2y_2$
C4	260,300, 340	400,480	500	$d_{xy} \longrightarrow dX^2y^2$

FT-IR spectra

FT-IR peaks as shown in Fig. 3 show the main peaks of uncoordinated ligand (L_1), which is C=C 1581.82 cm^{-1} and N=C 1621.82 cm^{-1}, and C-H 3052.58 cm^{-1}, stretching and also OH peak in 3292.48 cm^{-1} for $(L_1)^{22}$, Upon complexation, notable shifts in these bands are observed, The OH region with in the infrared spectrum with the metal compounds appear in the higher wavenumber in C_1 and C_3 in (3321.1, 3319.77) cm^{-1} and the shift of OH band to lower Wavenumber in (C_2 and C_4) (3200) cm^{-1} denoting the coordination within the atomic oxygen. The shift of C=N to lower wavenumber in (C_1-C_4): (1653.42, 1650, 1653.42, 1649.11) cm^{-1} respectively, which is indicated the coordination on C=N[16], also peaks in fingerprint reagent for C_1-C_4 between (500-700) cm^{-1}, showed the L-M peaks which indicate the complexation reaction[17], as shown in Table 2 and Figure 4.

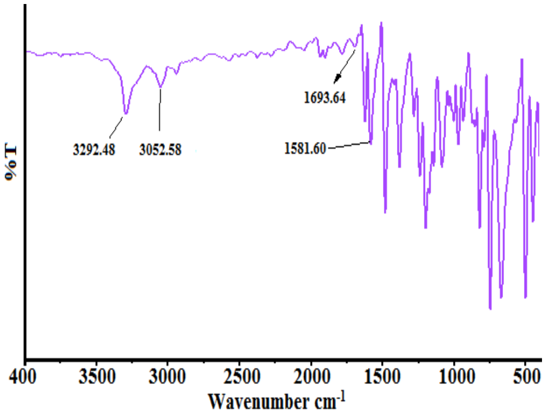


Fig. 3. FT-IR spectra of the Schiff base ligand (L_1)

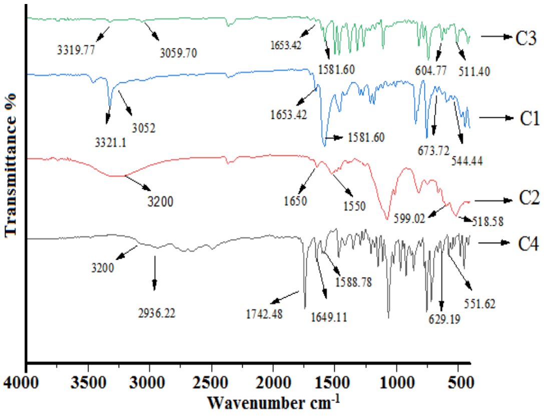


Fig. 4. FT-IR spectrum of Homo and Heterolepetic complexes (C_1- C_4)

Table 2: FT-IR Data of the ligand (L_1) and complexes of (C_1-C_4)

Compounds	OH	C=C	C=N	C-H	L-Metal	C=O
L_1	3292.48	1581.82	1693.64	3052.58	-	-
C_1	3321.1	1581.60	1653.42	3052	544.44,673.72	-
C_2	3200	1550	1650	-	518.58,599.02	-
C_3	3319.77	1581.60	1653.42	3059.70	511.40,604.7	-
C_4	3200	1588.78	1649.11	2936.22	551.62,629.19	1742.48

NMR spectra

^{1}H NMR (CDCl′, δ, ppm) peaks for (L_1) as shown in Fig. 5, a singlet is related to hydrogen of OH in 9.12 ppm also for HC=N give a peak in 8.72 ppm and also give (triplet, triplet, doublet, doublet, singlet) in the reagent between (6-9) ppm which are related to hydrogen of benzene ring[25]. The ligand gives a ^{13}C NMR peak at 158.7, which is related to carbon in C=N and peaks between (110-150), which is related to carbons of the benzene ring[26], as shown in Figure 6.

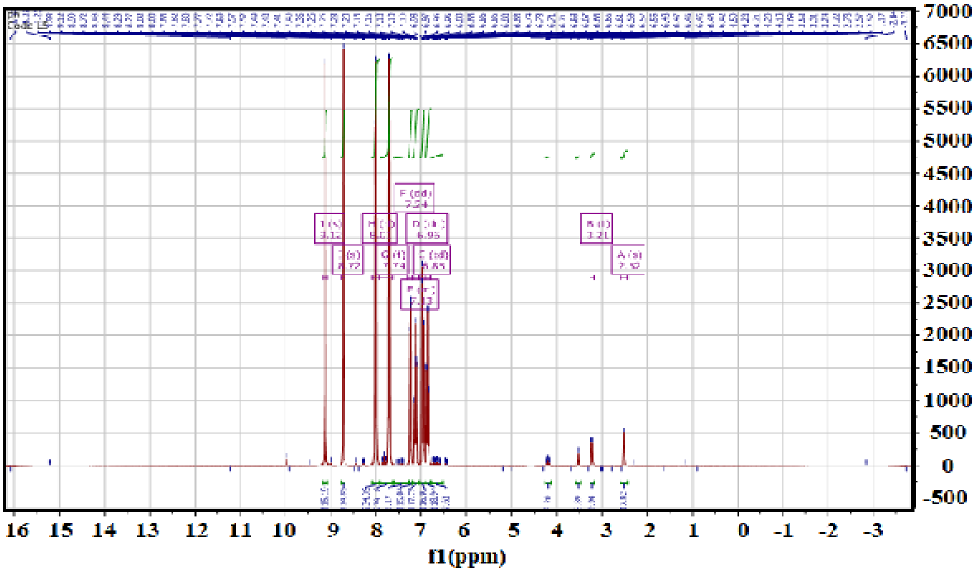


Fig. 5. ^{1}**H-NMR spectrum of Schiff base ligand (L_1)**

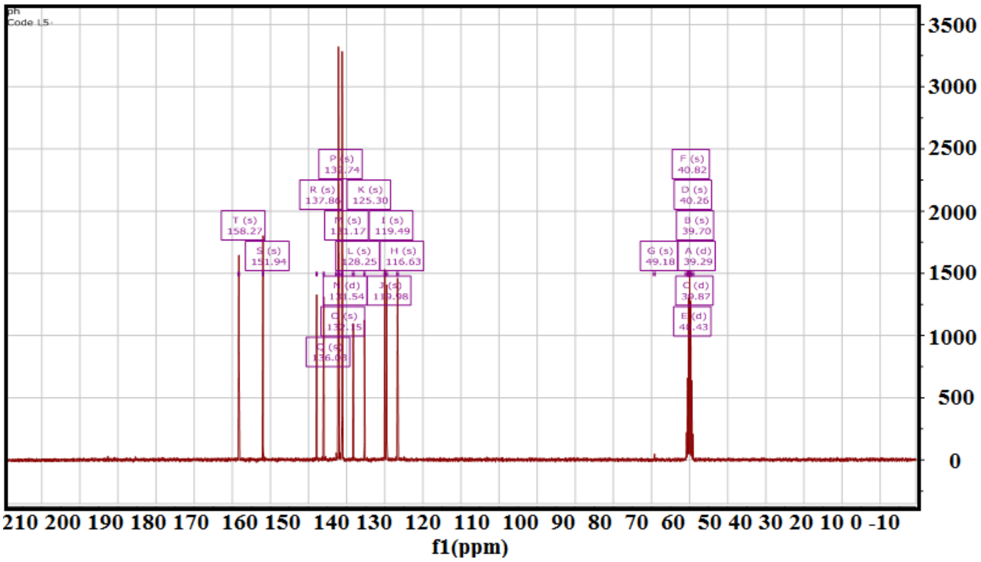


Fig. 6. ^{13}**C-NMR spectrum of Schiff base ligand (L_1)**

Elemental analysis

Emental analysis of (L_1) show amount of carbon, nitrogen, Hydrogen and also sulfur which amount of carbon is %55.326 and amount of hydrogen is %3.418 and amount of nitrogen is %5.032 and Sulphur is zero which all are close to theoretical amount, Theoretical amounts for all elements are {C (56.502), H (3.62), Br (28.91), N (5.07), S (0.00)}[27]. All measured elements are within 1% absolute of the theoretical values, confirming the sample's identity and high purity.

Magnetic moment

Magnetic moment of complexes determined by determination of mass susceptibility of complex compounds by using this equation: molar susceptibility (emu mol^{-1}), Xmeas = Xg × M.Wt, μeff = 2.83 (XpT1/2)(Bohr-magneton formula, T = 298 K), Calculated Bohr magneton for the complexes (C_1-C_4): (1.84), (1.7), (1.53), and (1.7) rspectvily[28], These values are all in the narrow range 1.5–1.8 BM, which is markedly value for one unpaired electron (1.73 BM).

Table 3: Analytical and physical properties of ligand (L_1) and their metals complexes (C_1-C_4)

Compounds	Molecular formula	Sstructure	μeff (B.M)	Color	Conductivity(S·cm^2/mol)	% Yield	Melting Point
L1	$C_{13}H_{10}NOBr$	-	-	Yello	-	%70	133-137
C1	$[Cu(L_1)_2Cl_2]$	Octahedral	1.84	Black	12.5	%60	>300
C2	$[Fe(L_1)_2Cl_2]Cl$	Octahedral	1.7	Grey	2.1	%85	>300
C3	$[Cu(L_1)(L_2)Cl_2]$	Octahedral	1.53	Green	6.1	%72	259-260
C4	$[Cu(L_1)(L_3)Cl_2]$	Octahedral	1.7	Green	13.8	%40	>300

DFT analysis

Quantum chemistry studies molecules' bond formations, molecular structures, energy levels, and chemical reactivity using computational methods. The three-dimensional arrangements of the compounds.

$[Cu(L_1)_2Cl_2]$ and $[Fe(L_1)_2Cl_2]Cl$ were created utilizing Gauss View 6.0 and further optimized utilizing the Gaussian 16 software suite. The study of geometry improvements and frequency calculations were performed in the gaseous phase utilizing the Becke, 3-parameter, Lee–Yang–Parr (B3LYP) operational.

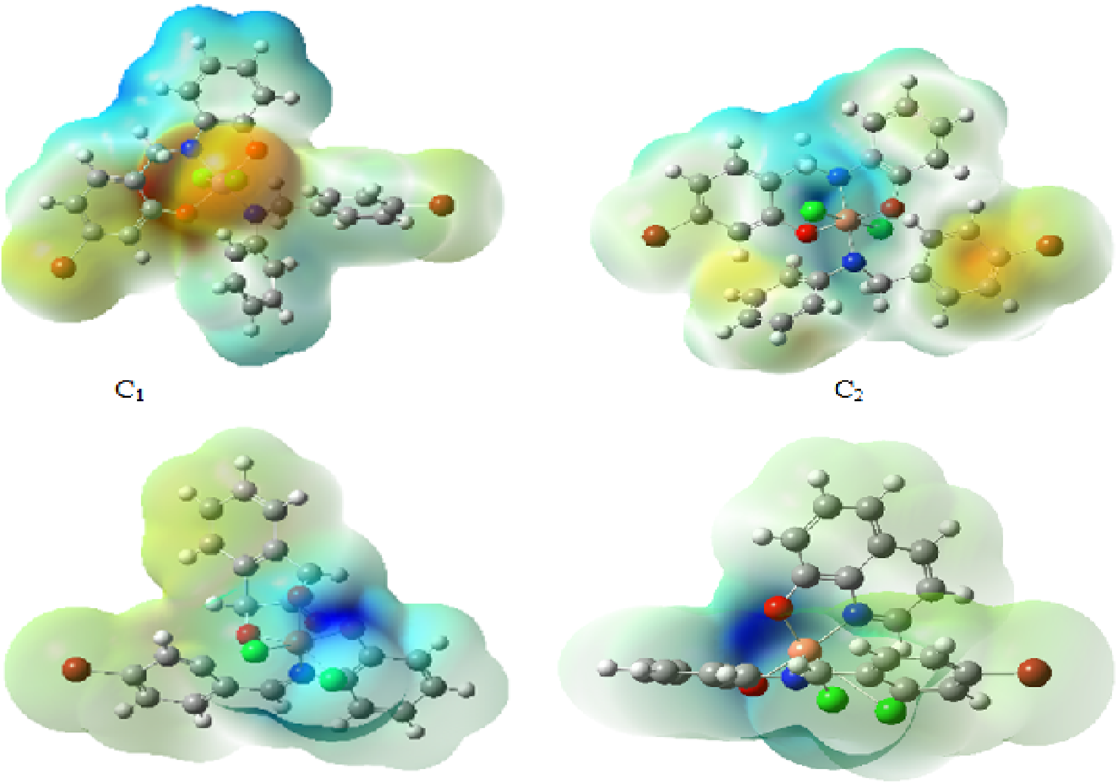
The B3LYP/6-311++G(d,p) A basic set was employed for nitrogen, carbon, oxygen, and hydrogen atoms.

The computed frequencies were subsequently optimized to confirm that the resulting structures were in the minimum energy state.

The Gauss View molecule imaging program was utilized to visualize the input data files as well as determine the HOMO–LUMO energies.[29]

Table 4: Appendix X. The electronic structure of the selected investigated compounds is described by quantumchemical parameters computed using DFT at the B3LYP/6-311++G(d,p) theoretical value in the gaseous phase

Quantum Chemical parameters	C1	C2	C3	C4
HOMO	-5.78895324	-6.31603805	-5.98297052	-4.27246191
LUMO	-5.22513303	-4.96716896	-4.23409384	-3.56578186
Ionization energy (eV) (I)	5.788953	6.316038	5.982971	4.272462
Electron Affinity (eV) (A)	5.225133	4.967169	4.234094	3.565782
Energy gap (eV) (ΔE)	0.56382	1.348869	1.748877	0.70668
Hardness (eV) (η)	0.28191	0.674435	0.874438	0.35334
Softness (eV)$^{-1}$ (σ)	3.54723	1.482724	1.143591	2.830135
Electronegativivty (eV) (χ)	5.507043	5.641604	5.108532	3.919122
Chemicalpotential (eV) (μ)	-5.50704	-5.6416	-5.10853	-3.91912
Electrophilicity (eV) (ω)	53.78935	23.59583	14.92221	21.73475
Nucleophilicity (eV)$^{-1}$ (ε)	0.018591	0.04238	0.067014	0.046009
Back-donation (eV) (ΔE_{BD})	-0.07048	-0.16861	-0.21861	-0.08834



C_1 C_2 C_3 C_4

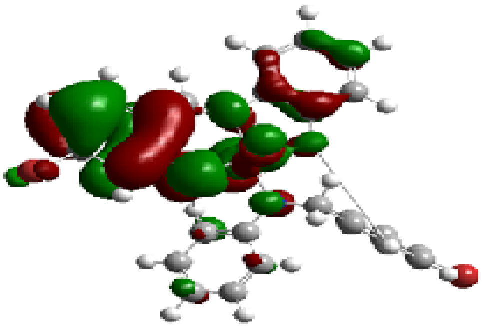
Fig. 7. Electrostatic potentials map of the selected compounds based on DFT at 6-311G++(d,p) basis set

HOMO and LOMO energy

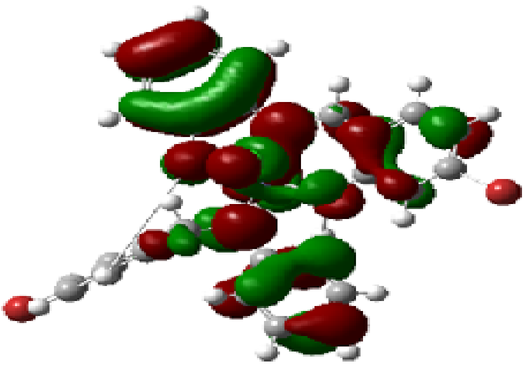
The HOMO energies: These values represent the energy of the highest occupied molecular orbital. The more negative the HOMO energy, the more stable the electrons in the compound. The HOMO energy for complexes (C_1-C_4):-5.788 Ev, -6.316 eV, -5.982 eV, -4.272 eV respectively.

The LUMO energies: These values represent the energy of the lowest unoccupied molecular orbital. The more negative the LUMO energy, the more difficult it is to excite an electron to this orbital. The LOMO energy for complexes (C_1-C_4):-5.225 ev, -4.967 Ev, -4.234 eV, -3.565 Ev respectively.

The Energy gaps: These values represent the difference between the HOMO and LUMO energies. A larger energy gap indicates a more stable compound. The energy gape for complexes (C_1-C_4): 0.56382 Ev, 1.348 Ev, 1.748 eV, 0.706 eV respectively the compound C_3 has the largest energy gap, indicating the most stable compound and C_2 has the most negative HOMO, LOMO and has the heights.



HOMO C_1



LOMO C_1

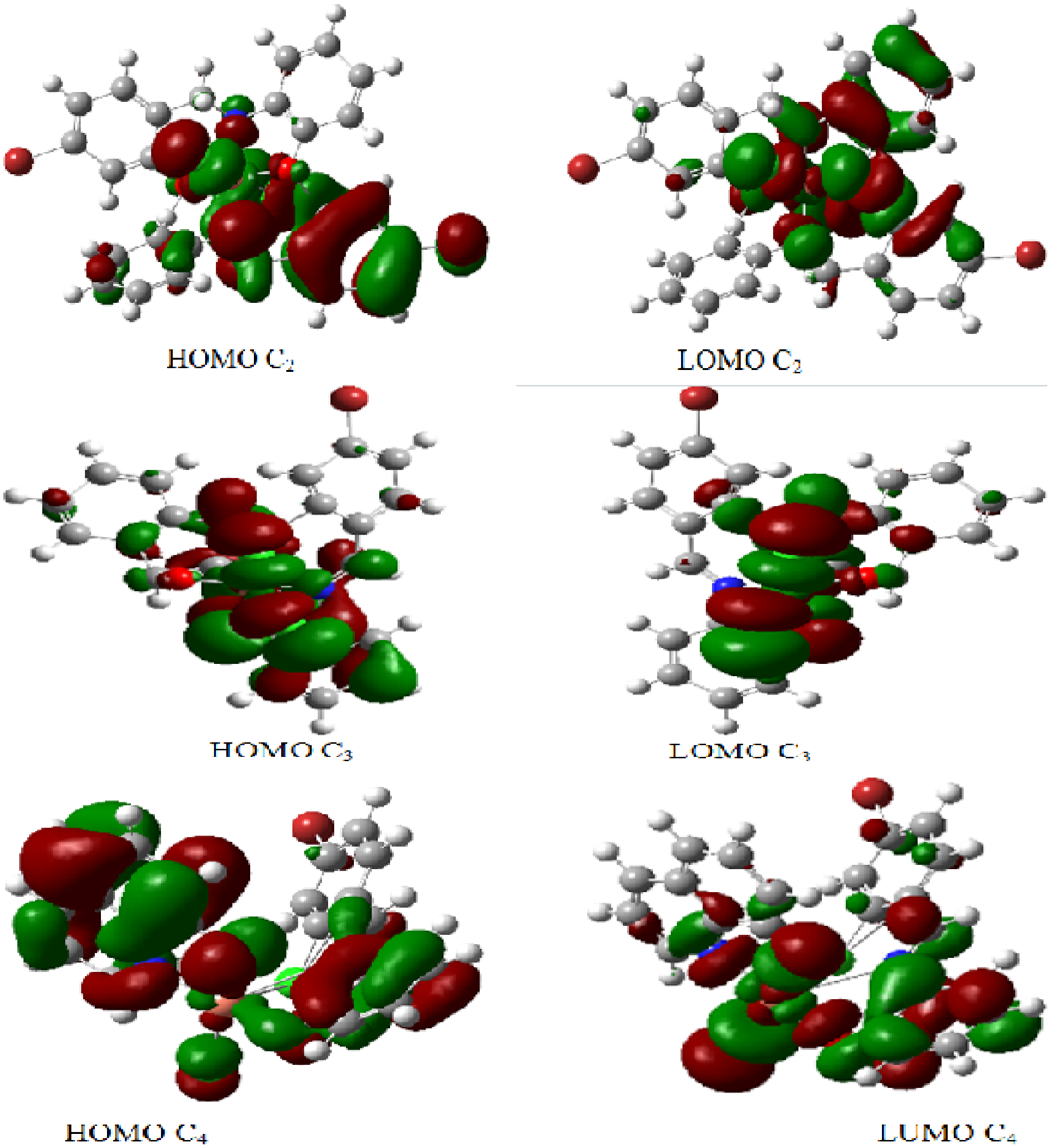


Fig. 8. HOMO and LUMO complexes of (C_1-C_4)

Antibacterial activity

The Schiff base ligand (L_1) and its corresponding complexes were evaluated for antibacterial activity against two bacterial strains: *Salmonella (Gram-negative)* and *Staphylococcus (Gram-positive)*[30], The disk diffusion method was employed, with the complexes tested in powder form, and the inhibition zones were measured in millimeters (mm) and the results listed in Table 5.

The homoleptic complexes (C_1 and C_2) exhibited strong antibacterial activity against both strains. The complex (C_1) displayed inhibition zones of 25 mm for *Salmonella* and 26 mm for *Staphylococcus*, while (C_2) showed even greater efficacy, with 30 mm inhibition zones for both bacteria. In contrast, the heteroleptic complexes (C_3 and C_4) demonstrated significantly reduced activity, suggesting that the homoleptic form of this ligand is more effective against these pathogens. Notably, the ligand (L_1) showed no inhibitory effect on either bacterial strain[31-33].

Table 5: Antibacterial activities of Ligand (L_1) and complexes (C_1-C_4) quantified in millimeters

Compounds	*Salmonella* (-)	*Staphylococcus* (+)
C_1	25 mm	26 mm
C_2	30 mm	30 mm
C_3	25 mm	25 mm
C_4	22 mm	14 mm
L_1	-	-

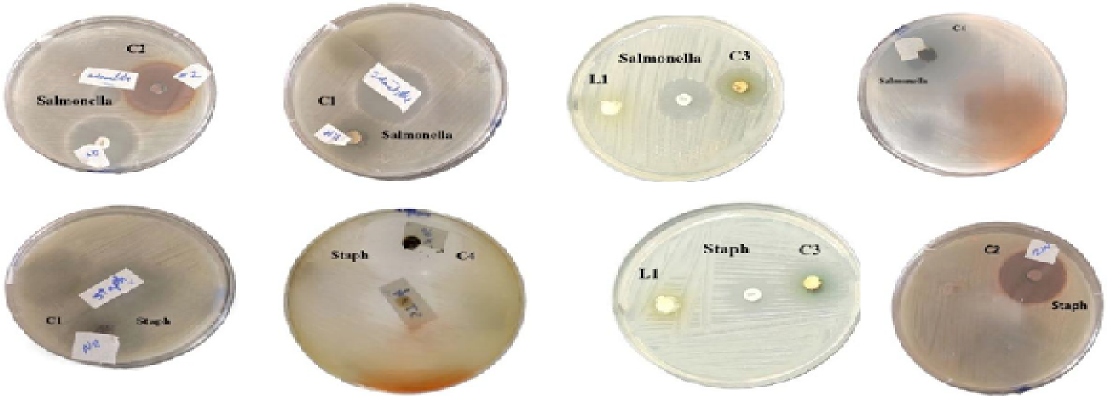


Fig. 9. Investigation antibacterial activity of Ligand (L_1) and their complexes (C1-C4)

Conclusion

This study confirms the successful synthesis of 2-(4-bromobenzylidene)aminophenol ligands and their Fe(II) and Cu(II) complexes, which were thoroughly characterized using spectral techniques and elemental analysies. The complexes were synthesized in a 1:1 metal to ligand (M:L) ratio and characterized by melting point, UV-Vis, FT-IR, and molar conductivity Octahedral geometry was established by magnetic and UV–Vis data, and molar conductance confirmed the complexes are non-electrolytes in solution. The Gauss View molecule imaging program was utilized to visualize the input data files and determine the HOMO–LUMO energies.

The synthesized complexes exhibited strong antibacterial activity against *Salmonella* and *Staphylococcus*. Notably, complexes (C_3 and C_4) showed reduced efficacy against both bacterial strains, suggesting that homoleptic complexes of this ligand possess enhanced antibacterial properties. In contrast, the free ligand (L_1) displayed no antibacterial activity.These findings underscore the importance of metal coordination in enhancing the biological activity of amide-based compounds, providing a foundation for further exploration of their therapeutic and pharmacological applications.

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Conflict of interest

The author declare that we have no conflict of interest.

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