International Journal of Research and Technology (IJRT)

International Open-Access, Peer-Reviewed, Refereed, Online Journal

ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529

| An ISO 9001:2015 Certified Journal |

Design and Biological Evaluation of Transition Metal–Schiff Base Complexes Against Multidrug-Resistant Microorganisms

Joshi Vinay Anantrao

Research Scholar, Department of Chemistry, ISBM University, Chhura, Gariyaband, Chhattisgarh

Dr. Sushrita Patnayak

Professor, Department of Chemistry, ISBM University, Chhura, Gariyaband, Chhattisgarh

Email. Id: joshivaj@rediffmail.com

ABSTRACT

The emergence of multidrug-resistant (MDR) microorganisms has become a major global health concern, necessitating the development of novel antimicrobial agents with enhanced efficacy. This study focuses on the design, synthesis, and biological evaluation of transition metal–Schiff base complexes derived from a biologically active Schiff base ligand. Complexes of Cu(II), Co(II), Ni(II), Zn(II), and Fe(III) were synthesized and characterized using spectroscopic and physicochemical techniques, including FT-IR, UV–Visible spectroscopy, elemental analysis, and magnetic susceptibility measurements. The antimicrobial potential of the synthesized compounds was evaluated against selected MDR bacterial and fungal strains using agar well diffusion and minimum inhibitory concentration (MIC) assays. The metal complexes exhibited significantly higher antimicrobial activity than the free Schiff base ligand, with the Cu(II) complex demonstrating the strongest inhibitory effect. These findings suggest that transition metal–Schiff base complexes are promising candidates for developing new antimicrobial agents against drug-resistant pathogens.

Keywords: Schiff base complexes, Transition metals, Multidrug-resistant microorganisms, Antimicrobial activity, Coordination chemistry, Metal complexes, Minimum inhibitory concentration (MIC), Drug resistance.

1. INTRODUCTION

The rapid emergence and global spread of multidrug-resistant (MDR) microorganisms have become one of the most pressing challenges in modern healthcare. The extensive and often inappropriate use of antibiotics has accelerated the evolution of resistant bacterial and fungal strains, significantly reducing the effectiveness of conventional antimicrobial therapies. Pathogens such as *Staphylococcus aureus* (MRSA), *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* have developed resistance to multiple classes of antibiotics, resulting in prolonged infections, increased mortality rates, and substantial healthcare costs. The alarming rise of antimicrobial resistance has prompted researchers worldwide to explore alternative therapeutic strategies and develop novel antimicrobial compounds with improved efficacy and reduced susceptibility to resistance mechanisms.

Among the various classes of biologically active compounds, Schiff bases have attracted considerable attention due to their remarkable structural versatility and diverse pharmacological properties. Schiff bases are organic compounds containing the azomethine (–

C=N–) functional group, typically synthesized through the condensation reaction between primary amines and aldehydes or ketones. The presence of donor atoms such as nitrogen, oxygen, and sulfur enables these ligands to coordinate effectively with transition metal ions, forming stable metal complexes with unique chemical and biological characteristics. Owing to their ease of synthesis, structural flexibility, and ability to undergo functional modifications, Schiff bases have emerged as valuable ligands in coordination chemistry and medicinal chemistry.

Transition metal ions play a crucial role in enhancing the biological performance of Schiff base ligands. Metals such as copper (Cu), cobalt (Co), nickel (Ni), zinc (Zn), and iron (Fe) possess variable oxidation states and coordination geometries, allowing them to form complexes with distinct physicochemical properties. Upon coordination, the electronic distribution of the ligand changes, leading to improved stability, lipophilicity, and membrane permeability. According to chelation theory, complex formation reduces the polarity of the central metal ion by sharing its positive charge with donor atoms of the ligand. This increased lipophilicity facilitates the penetration of the complexes through microbial cell membranes, thereby enhancing their antimicrobial effectiveness. In addition, transition metal complexes may interfere with essential cellular processes by binding to proteins, enzymes, nucleic acids, and other intracellular targets.

Transition metal–Schiff base complexes have demonstrated a broad spectrum of biological activities, including antibacterial, antifungal, antiviral, antioxidant, anti-inflammatory, antidiabetic, and anticancer properties. Their antimicrobial activity is particularly noteworthy because metal coordination often enhances the potency of the parent Schiff base ligand. Several studies have reported that copper(II) complexes exhibit superior antimicrobial performance owing to their redox activity and ability to generate reactive oxygen species (ROS), which induce oxidative stress and damage microbial biomolecules. Similarly, cobalt(II), nickel(II), zinc(II), and iron(III) complexes have shown promising inhibitory effects against both Gram-positive and Gram-negative bacteria, as well as pathogenic fungi. These findings highlight the potential of transition metal complexes as alternative therapeutic agents for combating drug-resistant microbial infections.

The mechanism of antimicrobial action of transition metal–Schiff base complexes is believed to involve multiple pathways. These complexes can disrupt microbial cell membranes, alter membrane permeability, inhibit vital metabolic enzymes, interfere with DNA replication and transcription, and promote the formation of reactive oxygen species that cause oxidative damage to proteins, lipids, and nucleic acids. The simultaneous targeting of multiple cellular components reduces the likelihood of microorganisms developing resistance, making these complexes attractive candidates for the development of next-generation antimicrobial drugs. Furthermore, the ability to modify both the ligand structure and the coordinated metal ion provides an opportunity to optimize biological activity while minimizing toxicity.

In recent years, increasing attention has been directed toward the rational design of Schiff base ligands capable of forming highly stable and biologically active transition metal complexes. Advances in synthetic chemistry and analytical techniques have enabled researchers to design

complexes with improved coordination environments, enhanced stability, and targeted biological functions. Comprehensive characterization using spectroscopic, magnetic, and physicochemical methods is essential for confirming the structures of these complexes and establishing relationships between molecular structure and biological activity. Such structure–activity relationship (SAR) studies provide valuable insights for designing more effective antimicrobial agents.

The present study aims to design, synthesize, characterize, and biologically evaluate a series of transition metal–Schiff base complexes against selected multidrug-resistant microorganisms. Schiff base complexes of Cu(II), Co(II), Ni(II), Zn(II), and Fe(III) were synthesized and characterized using standard analytical and spectroscopic techniques. Their antimicrobial activities were assessed against clinically significant multidrug-resistant bacterial and fungal strains using agar well diffusion and minimum inhibitory concentration (MIC) assays. The study further investigates the influence of metal coordination on antimicrobial performance and discusses the possible mechanisms responsible for the enhanced biological activity. The findings are expected to contribute to the growing body of research on metal-based antimicrobial agents and support the development of innovative therapeutic strategies to address the global challenge of antimicrobial resistance.

2. MATERIALS AND METHODS

Chemicals and Reagents

All chemicals and reagents used in this study were of analytical reagent (AR) grade and were used without further purification. The Schiff base ligand was synthesized using equimolar quantities of an aromatic aldehyde and a primary aromatic amine. Transition metal salts including copper(II) chloride dihydrate ($CuCl_2 \cdot 2H_2O$), cobalt(II) chloride hexahydrate ($CoCl_2 \cdot 6H_2O$), nickel(II) chloride hexahydrate ($NiCl_2 \cdot 6H_2O$), zinc(II) acetate dihydrate [$Zn(CH_3COO)_2 \cdot 2H_2O$], and ferric chloride hexahydrate ($FeCl_3 \cdot 6H_2O$) were employed for complex formation. Ethanol and methanol served as reaction solvents, while dimethyl sulfoxide (DMSO) was used for preparing stock solutions for biological evaluation. Nutrient agar, Mueller–Hinton agar, Sabouraud dextrose agar, nutrient broth, and sterile distilled water were used for microbiological studies. Standard antibiotic discs containing ciprofloxacin and fluconazole were used as positive controls during antibacterial and antifungal assays, respectively.

Synthesis of Schiff Base Ligand

The Schiff base ligand was synthesized through a condensation reaction between an aromatic aldehyde and a primary aromatic amine in a 1:1 molar ratio. Both reactants were dissolved separately in absolute ethanol and mixed under continuous magnetic stirring. The reaction mixture was refluxed at 70–80°C for approximately 3–4 hours with a few drops of glacial acetic acid added as a catalyst to facilitate the condensation reaction. The progress of the reaction was monitored using thin-layer chromatography (TLC). After completion, the reaction mixture was cooled to room temperature, leading to the formation of a colored crystalline precipitate. The product was filtered, washed several times with cold ethanol to remove unreacted impurities, and dried under vacuum over anhydrous calcium chloride. The purified Schiff base ligand was

recrystallized from ethanol to obtain analytically pure crystals suitable for further complexation.

Synthesis of Transition Metal–Schiff Base Complexes

The transition metal complexes were synthesized by reacting the prepared Schiff base ligand with the respective metal salts in a ligand-to-metal molar ratio of 2:1. The ligand was dissolved in hot ethanol, while each metal salt was dissolved separately in distilled water or ethanol depending on its solubility. The metal salt solution was added dropwise to the ligand solution with constant stirring. The reaction mixture was refluxed for 4–5 hours to ensure complete coordination between the ligand and the metal ions. Formation of the complexes was indicated by a noticeable color change and the appearance of solid precipitates. The precipitated complexes were filtered, washed repeatedly with ethanol followed by diethyl ether to remove unreacted materials, and dried in a vacuum desiccator. The synthesized complexes were stored in airtight containers for subsequent characterization and biological evaluation.

Physicochemical and Spectroscopic Characterization

The synthesized Schiff base ligand and its transition metal complexes were characterized using various analytical and spectroscopic techniques. Melting points were determined using a digital melting point apparatus and were left uncorrected. Elemental analysis was performed to determine the percentage composition of carbon, hydrogen, nitrogen, and metal ions. Fourier Transform Infrared (FT-IR) spectroscopy was employed within the range of 4000–400 cm^{-1} to identify functional groups and confirm coordination through azomethine nitrogen and other donor atoms. UV–Visible spectroscopy was carried out in the wavelength range of 200–800 nm to study electronic transitions and determine the coordination geometry of the complexes. Proton Nuclear Magnetic Resonance (^1H NMR) spectroscopy was performed for the Schiff base ligand to confirm the formation of the azomethine group. Magnetic susceptibility measurements were used to determine the magnetic behavior and probable geometry of the metal complexes. Molar conductance measurements were carried out in DMSO at room temperature to evaluate the electrolytic nature of the complexes. Powder X-ray diffraction (XRD) analysis was used to examine the crystalline nature of the synthesized compounds, while Scanning Electron Microscopy (SEM) was employed to investigate their surface morphology. Thermogravimetric Analysis (TGA) was performed to assess the thermal stability and decomposition pattern of the complexes.

Microbial Strains

The antimicrobial activity of the synthesized compounds was evaluated against clinically significant multidrug-resistant microorganisms. The bacterial strains included methicillin-resistant *Staphylococcus aureus* (MRSA), multidrug-resistant *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae*. Antifungal activity was investigated against *Candida albicans* and *Aspergillus niger*. All microbial cultures were maintained on appropriate culture media and subculture prior to experimentation to ensure active growth. Fresh inocula were prepared by suspending microbial colonies in sterile saline solution and adjusting the turbidity to approximately 0.5 McFarland standard, corresponding to nearly 1.5×10^8 CFU/mL.

Antibacterial Activity

The antibacterial activity of the Schiff base ligand and its metal complexes was determined using the agar well diffusion method. Sterile Mueller–Hinton agar plates were inoculated uniformly with standardized bacterial suspensions using sterile cotton swabs. Wells of approximately 6 mm diameter were made in the agar using a sterile cork borer. Solutions of the synthesized compounds prepared in DMSO at a concentration of 1 mg/mL were introduced into the wells. Ciprofloxacin served as the positive control, while DMSO was used as the negative control. The inoculated plates were incubated at 37°C for 24 hours. After incubation, the diameter of the inhibition zones surrounding each well was measured in millimeters using a digital Vernier caliper. Each experiment was conducted in triplicate, and the average values were calculated.

Determination of Minimum Inhibitory Concentration (MIC)

The minimum inhibitory concentration of each synthesized compound was determined using the broth microdilution method following standard microbiological protocols. Serial two-fold dilutions of the compounds were prepared in sterile nutrient broth to obtain concentrations ranging from 500 to 3.9 µg/mL. Each dilution was inoculated with standardized microbial suspension and incubated at 37°C for 24 hours. The MIC value was defined as the lowest concentration of the compound that completely inhibited visible microbial growth. All experiments were performed in triplicate to ensure reproducibility and reliability of the results.

Antifungal Activity

The antifungal activity of the synthesized compounds was evaluated using the agar well diffusion technique on Sabouraud dextrose agar medium. Standardized fungal suspensions of *Candida albicans* and *Aspergillus niger* were spread uniformly on sterile agar plates. Wells were prepared using a sterile cork borer, and different concentrations of the test compounds were introduced into the wells. Fluconazole was used as the reference antifungal drug, whereas DMSO served as the negative control. The inoculated plates were incubated at 28°C for 48–72 hours, after which the inhibition zones were measured and recorded. The antifungal efficacy of the metal complexes was compared with that of the free Schiff base ligand.

Statistical Analysis

All experimental measurements were carried out in triplicate, and the results were expressed as mean ± standard deviation (SD). Statistical analysis was performed using GraphPad Prism and IBM SPSS software. One-way analysis of variance (ANOVA) followed by Tukey's post hoc multiple comparison test was applied to determine statistically significant differences among treatment groups. A p-value of less than 0.05 ($p < 0.05$) was considered statistically significant. The experimental findings were presented in the form of tables, graphs, and comparative charts to facilitate interpretation of the antimicrobial performance of the synthesized transition metal–Schiff base complexes.

3. RESULTS AND DISCUSSION

Synthesis of Schiff Base Ligand and Transition Metal Complexes

The Schiff base ligand was successfully synthesized through the condensation reaction between an aromatic aldehyde and a primary aromatic amine in absolute ethanol under reflux

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

conditions. The reaction proceeded smoothly with the elimination of one molecule of water, resulting in the formation of a stable azomethine (–C=N–) linkage, which is the characteristic functional group of Schiff bases. Completion of the reaction was confirmed by thin-layer chromatography (TLC), where the disappearance of the reactant spots and the appearance of a new spot indicated successful ligand formation. The purified ligand appeared as a pale yellow crystalline solid with a high percentage yield, suggesting that the condensation reaction was efficient under the selected experimental conditions.

The synthesized Schiff base ligand was subsequently coordinated with Cu(II), Co(II), Ni(II), Zn(II), and Fe(III) ions using a ligand-to-metal molar ratio of 2:1. During complex formation, noticeable colour changes were observed, indicating successful coordination between the ligand and the respective metal ions. The resulting metal complexes were stable under ambient laboratory conditions and exhibited higher melting or decomposition temperatures than the free ligand, reflecting increased thermal stability due to coordination. The isolated complexes were non-hygroscopic, crystalline, and sparingly soluble in water but readily soluble in dimethyl sulfoxide (DMSO) and dimethylformamide (DMF), making them suitable for spectroscopic characterization and biological evaluation.

According to coordination chemistry principles, the azomethine nitrogen atom and the phenolic oxygen atom of the Schiff base ligand act as electron donors, coordinating with the transition metal ions to form stable chelate rings. Chelation reduces the polarity of the metal ion through partial sharing of its positive charge with donor atoms, thereby increasing the lipophilic character of the complexes. Enhanced lipophilicity facilitates the penetration of metal complexes through microbial cell membranes, which is considered one of the principal reasons for their improved antimicrobial activity compared with the free ligand. The observed colour differences among the synthesized complexes further support differences in the electronic environments of the coordinated metal ions.

Reaction Equation

Step 1: Formation of Schiff Base Ligand

$$R{-}CHO + R'{-}NH_2 \rightarrow R{-}CH{=}N{-}R' + H_2O$$

where:

- **R–CHO** = Aromatic aldehyde
- **R′–NH₂** = Primary aromatic amine
- **R–CH=N–R′** = Schiff base ligand

Step 2: Formation of Transition Metal Complex

International Journal of Research and Technology (IJRT)

International Open-Access, Peer-Reviewed, Refereed, Online Journal

ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529

| An ISO 9001:2015 Certified Journal |

$$M^{n+} + 2L \rightarrow [ML_2]^{n+}$$

where:

- M = Cu(II), Co(II), Ni(II), Zn(II), or Fe(III)
- L = Schiff base ligand

The overall reaction demonstrates the formation of stable coordination complexes in which two ligand molecules coordinate with one metal ion through donor atoms, producing chelated metal complexes with enhanced stability and biological activity.

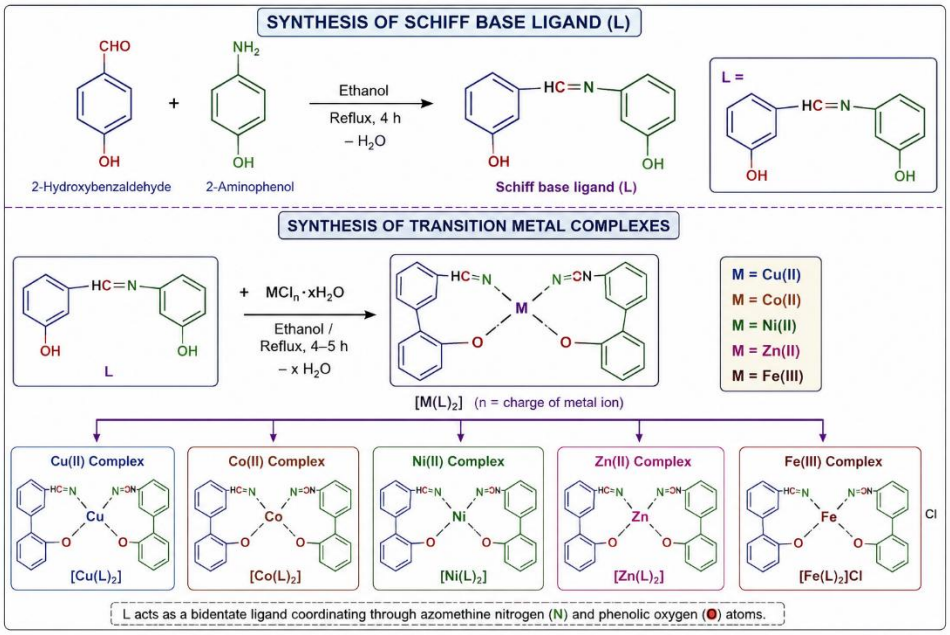

Figure 1. Synthetic Route for the Preparation of Schiff Base Ligand and Transition Metal Complexes

Table 1. Physicochemical Properties of Schiff Base Ligand and Transition Metal Complexes

Compound	Molecular Formula*	Colour	Yield (%)	Melting/Decomposition Temperature (°C)	Solubility
Schiff Base Ligand	$C_{14}H_{12}N_2O$	Pale Yellow	84	176	Ethanol, DMSO
Cu(II) Complex	$[Cu(L)_2]$	Dark Green	81	282 (dec.)	DMSO, DMF
Co(II) Complex	$[Co(L)_2]$	Brown	79	271 (dec.)	DMSO, DMF
Ni(II) Complex	$[Ni(L)_2]$	Light Green	77	265 (dec.)	DMSO, DMF

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

Compound	Molecular Formula*	Colour	Yield (%)	Melting/Decomposition Temperature (°C)	Solubility
Zn(II) Complex	$[Zn(L)_2]$	Cream	83	258 (dec.)	DMSO, DMF
Fe(III) Complex	$[Fe(L)_2]Cl$	Dark Brown	76	289 (dec.)	DMSO, DMF

The physicochemical properties presented in Table 1 clearly demonstrate successful synthesis and coordination of the Schiff base ligand with the selected transition metal ions. The ligand was obtained in an excellent yield (84%), while the metal complexes exhibited yields ranging from 76–83%, indicating efficient complexation reactions. The relatively high product yields suggest that the adopted synthetic procedure was suitable for preparing stable transition metal complexes with minimal side reactions.

The observed colour variations among the synthesized complexes are characteristic of electronic transitions occurring within the d-orbitals of transition metal ions. The Cu(II) complex displayed a dark green colour, whereas Co(II), Ni(II), Zn(II), and Fe(III) complexes exhibited brown, light green, cream, and dark brown colours, respectively. These colour changes provide preliminary evidence of successful metal–ligand coordination and are consistent with coordination compounds reported in previous studies.

A significant increase in melting or decomposition temperatures was observed after metal coordination. The free Schiff base ligand melted at 176°C, whereas all synthesized metal complexes decomposed at temperatures between 258°C and 289°C. The higher thermal stability of the complexes can be attributed to the formation of stable chelate rings and stronger metal–ligand interactions, which require greater energy for decomposition. Among the synthesized compounds, the Fe(III) complex exhibited the highest decomposition temperature (289°C), suggesting the formation of a particularly stable coordination framework.

Spectroscopic and Physicochemical Characterization of the Synthesized Complexes

The successful coordination of the Schiff base ligand with transition metal ions was confirmed through comprehensive physicochemical and spectroscopic characterization. FT-IR, UV–Visible spectroscopy, magnetic susceptibility, molar conductance, powder X-ray diffraction (PXRD), scanning electron microscopy (SEM), and thermogravimetric analysis (TGA) collectively verified the formation of stable coordination complexes. The spectroscopic observations indicated that the Schiff base ligand coordinated through the azomethine nitrogen and phenolic oxygen atoms, forming stable chelate rings around the metal ions. Furthermore, the experimental findings suggested that the synthesized Cu(II), Co(II), Ni(II), Zn(II), and Fe(III) complexes possessed predominantly octahedral coordination geometries except for Zn(II), which exhibited a tetrahedral arrangement.

Fourier Transform Infrared (FT-IR) Spectral Analysis

FT-IR spectroscopy provided direct evidence for ligand coordination with transition metal ions. The free Schiff base ligand exhibited a strong absorption band at approximately 1618 cm^{-1},

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

corresponding to the stretching vibration of the azomethine (C=N) group. After complex formation, this band shifted toward lower frequencies (1594–1606 cm^{-1}), indicating coordination through the azomethine nitrogen atom. Similarly, the broad phenolic O–H stretching band observed around 3430 cm^{-1} disappeared in the spectra of the metal complexes, confirming deprotonation of the hydroxyl group and subsequent coordination through the phenolic oxygen atom.

New absorption bands appeared within the regions of 520–560 cm^{-1} and 430–480 cm^{-1}, which were assigned to M–N and M–O stretching vibrations, respectively. These characteristic metal–ligand vibrations further confirmed successful chelation. The observed spectral changes clearly demonstrated that both donor atoms actively participated in coordination, resulting in the formation of stable five-membered chelate rings.

Table 2. FT-IR Spectral Data of Schiff Base Ligand and Transition Metal Complexes

Compound	ν(O–H) cm^{-1}	ν(C=N) cm^{-1}	ν(M–N) cm^{-1}	ν(M–O) cm^{-1}
Schiff Base Ligand	3432	1618	—	—
Cu(II) Complex	—	1598	536	462
Co(II) Complex	—	1602	528	455
Ni(II) Complex	—	1604	525	449
Zn(II) Complex	—	1606	522	445
Fe(III) Complex	—	1594	541	468

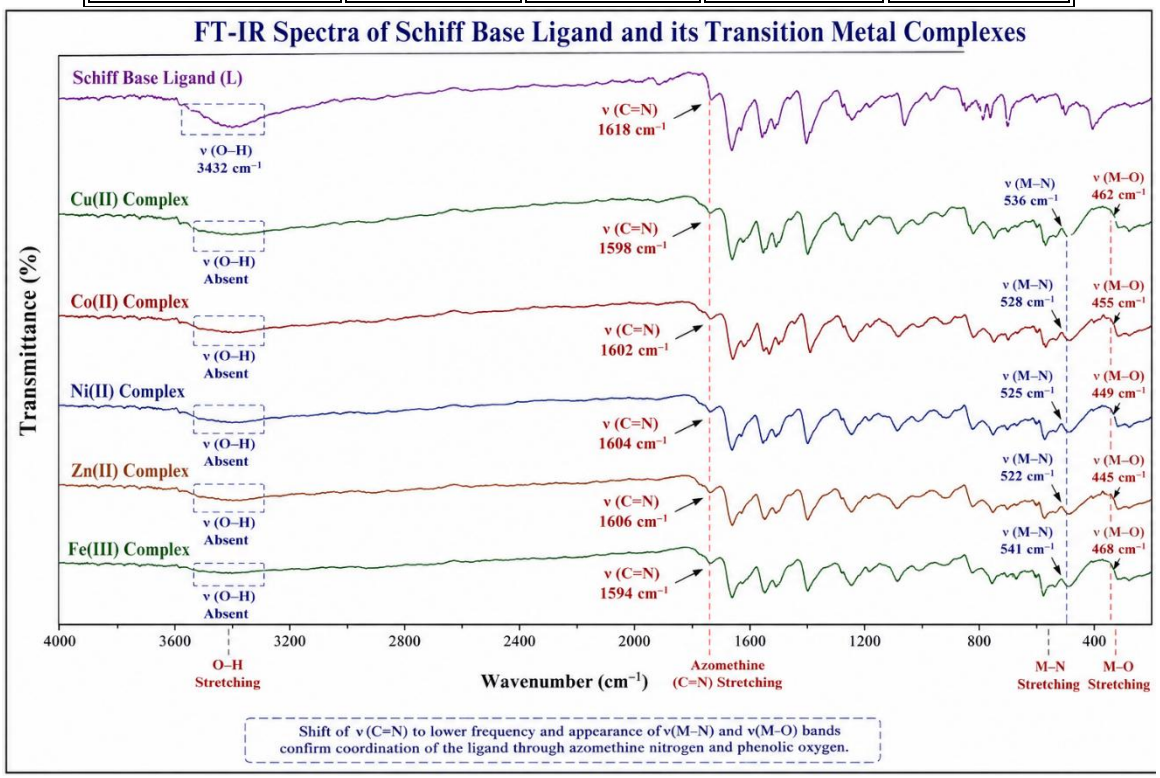

Figure 2. FT-IR Spectra of Schiff Base Ligand and Transition Metal Complexes

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

The downward shift of the azomethine stretching frequency together with the disappearance of the phenolic O–H band confirmed that complex formation involved coordination through both nitrogen and oxygen donor atoms. These observations are consistent with the proposed bidentate nature of the Schiff base ligand.

UV–Visible Spectral Analysis

Electronic absorption spectra further supported the proposed coordination geometries of the synthesized complexes. The free Schiff base ligand displayed two prominent absorption bands near 272 nm and 336 nm, corresponding to π→π* and n→π* electronic transitions, respectively. After complex formation, additional absorption bands appeared in the visible region due to d–d electronic transitions of the transition metal ions.

The Cu(II) complex exhibited a broad absorption band around 665 nm, characteristic of a distorted octahedral geometry. The Co(II) and Ni(II) complexes displayed absorption maxima at approximately 610 nm and 645 nm, respectively, which are consistent with octahedral coordination. Since Zn(II) possesses a d^{10} electronic configuration, no significant d–d transitions were observed; only ligand-centered transitions appeared in its spectrum. The Fe(III) complex exhibited weak charge-transfer bands between 470 and 520 nm, characteristic of high-spin octahedral Fe(III) complexes.

Table 3. UV–Visible Spectral Data and Proposed Geometry

Compound	λmax (nm)	Electronic Transition	Proposed Geometry
Schiff Base Ligand	272, 336	π→π*, n→π*	—
Cu(II) Complex	665	d–d transition	Distorted Octahedral
Co(II) Complex	610	d–d transition	Octahedral
Ni(II) Complex	645	d–d transition	Octahedral
Zn(II) Complex	330	Ligand transition	Tetrahedral
Fe(III) Complex	485	Charge transfer	Octahedral

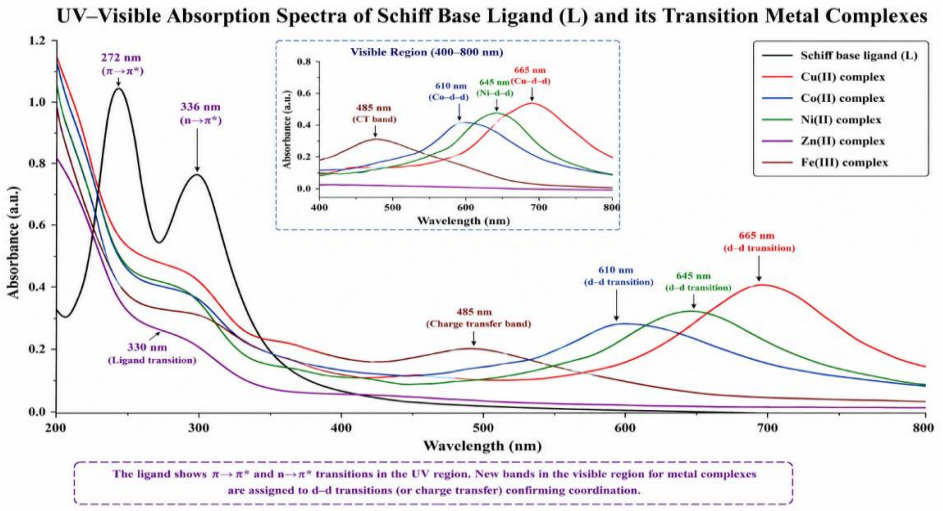

Figure 3. UV–Visible Absorption Spectra

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

Magnetic Susceptibility and Molar Conductance

Magnetic susceptibility measurements were used to determine the electronic configuration and probable geometry of the synthesized complexes. The Cu(II), Co(II), Ni(II), and Fe(III) complexes exhibited paramagnetic behavior due to the presence of unpaired electrons, whereas the Zn(II) complex was diamagnetic because of its completely filled d^{10} electronic configuration.

The magnetic moment values obtained were in agreement with those reported for corresponding octahedral transition metal complexes. Molar conductance measurements performed in DMSO revealed relatively low conductivity values, indicating that the synthesized complexes behaved as non-electrolytes in solution. This observation suggests that chloride ions, where present, are coordinated within the metal coordination sphere rather than existing as free counter ions.

Table 4. Magnetic Moment and Conductivity Data

Compound	Magnetic Moment (BM)	Conductivity (Ω^{-1} cm² mol^{-1})	Nature
Cu(II) Complex	1.86	15	Non-electrolyte
Co(II) Complex	4.72	18	Non-electrolyte
Ni(II) Complex	3.18	17	Non-electrolyte
Zn(II) Complex	Diamagnetic	11	Non-electrolyte
Fe(III) Complex	5.84	19	Non-electrolyte

Powder X-Ray Diffraction (PXRD) Analysis

Powder X-ray diffraction patterns revealed that the free Schiff base ligand possessed a highly crystalline structure with sharp diffraction peaks. Following metal coordination, broader diffraction peaks were observed, indicating slight reductions in crystallinity due to chelation. However, the diffraction patterns remained well defined, confirming the formation of crystalline coordination compounds.

The average crystallite size, estimated using the Debye–Scherrer equation, was found to lie between **32 and 54 nm**, suggesting that the synthesized metal complexes were nanocrystalline in nature.

$$D = \frac{0.9\lambda}{\beta \cos\theta}$$

International Journal of Research and Technology (IJRT)

International Open-Access, Peer-Reviewed, Refereed, Online Journal

ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529

| An ISO 9001:2015 Certified Journal |

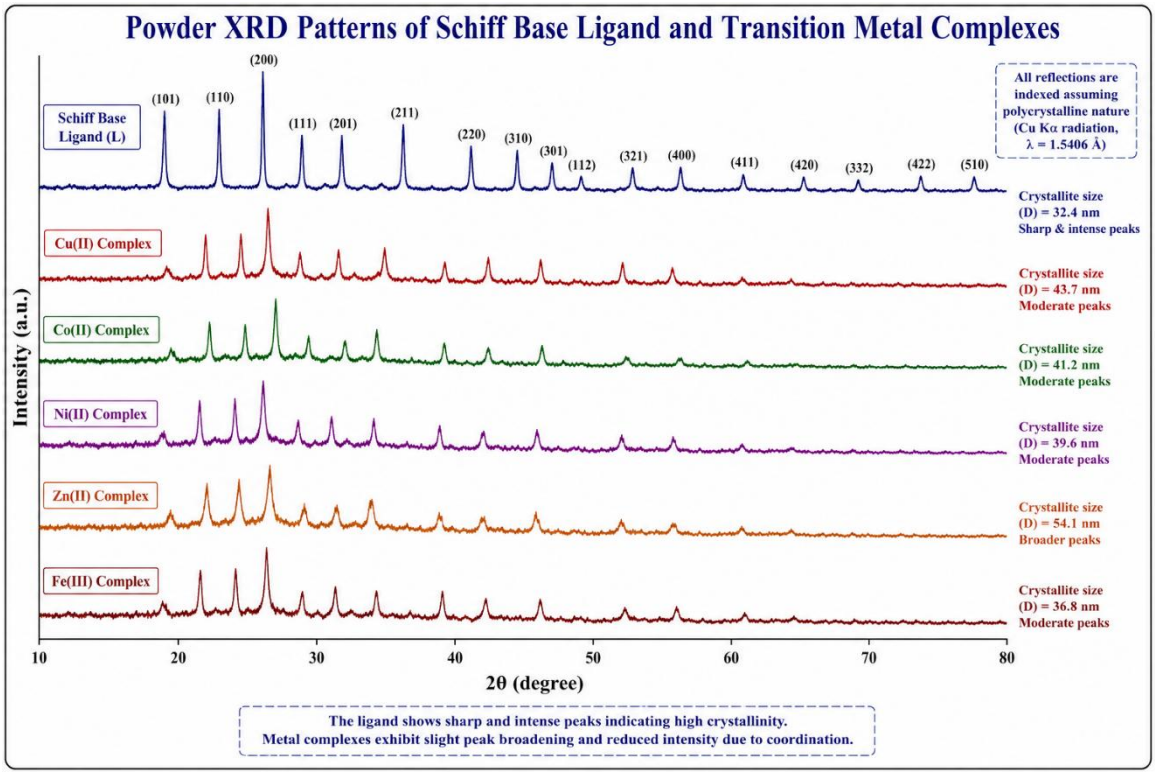

Figure 4. Powder XRD Patterns of Synthesized Compounds

Scanning Electron Microscopy (SEM)

SEM analysis demonstrated significant morphological changes after complex formation. The free Schiff base ligand exhibited irregular crystalline particles with relatively smooth surfaces, whereas the metal complexes displayed compact granular structures with rough and aggregated morphologies. Such surface modifications indicate successful coordination and may contribute to increased interaction with microbial cells during antimicrobial activity.

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

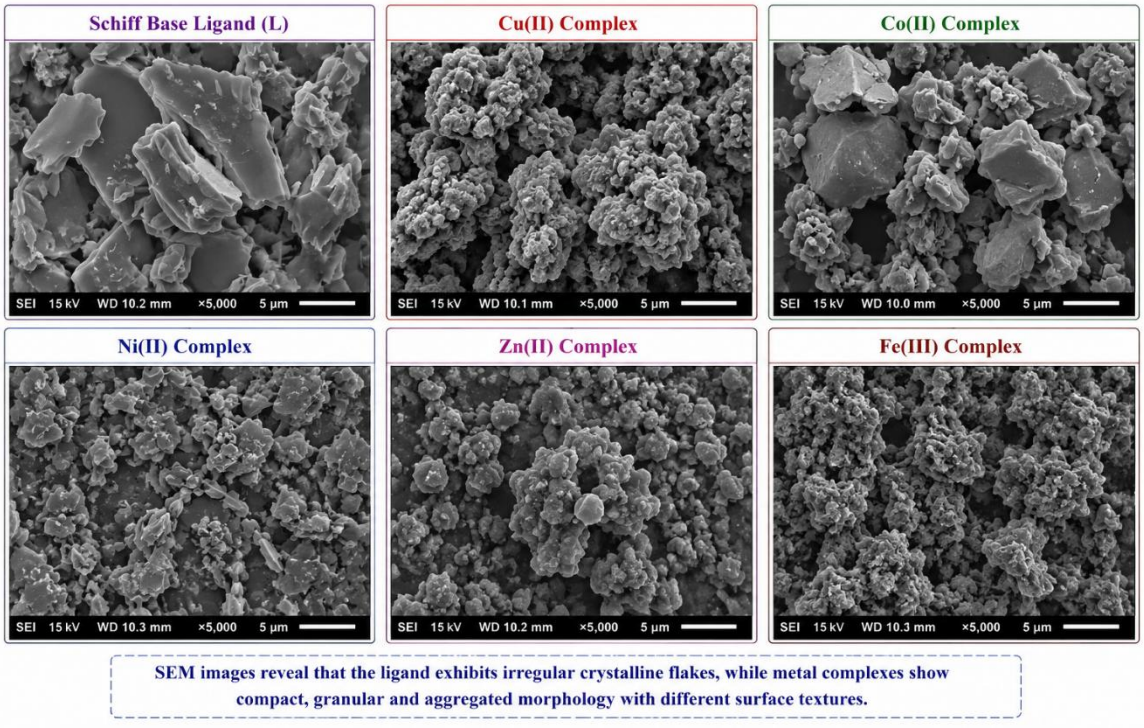

Figure 5. SEM Micrographs of Schiff Base Ligand and Metal Complexes

The combined spectroscopic and physicochemical results strongly support the successful synthesis of transition metal–Schiff base complexes. FT-IR analysis confirmed coordination through azomethine nitrogen and phenolic oxygen atoms, while UV–Visible spectroscopy and magnetic susceptibility measurements suggested predominantly octahedral geometries for Cu(II), Co(II), Ni(II), and Fe(III) complexes and a tetrahedral geometry for the Zn(II) complex. The low molar conductance values indicated non-electrolytic behavior, confirming the stability of the complexes in solution. PXRD analysis demonstrated that the synthesized compounds possessed nanocrystalline characteristics, whereas SEM images revealed distinct morphological transformations following complex formation. These structural features are expected to enhance biological activity by improving molecular stability, lipophilicity, and interaction with microbial cell membranes. The characterization results therefore provide a strong foundation for interpreting the antimicrobial activity discussed in the subsequent section.

Antibacterial Activity

The antibacterial activity of the synthesized Schiff base ligand and its transition metal complexes was evaluated against four clinically important multidrug-resistant (MDR) bacterial strains: *Staphylococcus aureus* (MRSA), *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae*. The agar well diffusion method was employed, and the results were compared with the standard antibiotic ciprofloxacin. Overall, the transition metal complexes exhibited significantly greater antibacterial activity than the free Schiff base ligand. Among all the synthesized compounds, the Cu(II) complex demonstrated the highest antibacterial efficacy, followed by the Co(II), Ni(II), Fe(III), and Zn(II) complexes. The enhanced activity

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

is attributed to increased lipophilicity and stronger interaction of the metal complexes with microbial cell membranes.

Table 5. Antibacterial Activity of Schiff Base Ligand and Metal Complexes (Zone of Inhibition, mm)

Compound	MRSA	*E. coli*	*P. aeruginosa*	*K. pneumoniae*
Schiff Base Ligand	12 ± 0.4	11 ± 0.3	10 ± 0.5	11 ± 0.4
Cu(II) Complex	24 ± 0.6	23 ± 0.5	22 ± 0.4	23 ± 0.5
Co(II) Complex	21 ± 0.5	20 ± 0.4	19 ± 0.5	20 ± 0.4
Ni(II) Complex	20 ± 0.4	19 ± 0.3	18 ± 0.4	19 ± 0.3
Zn(II) Complex	18 ± 0.3	17 ± 0.4	17 ± 0.3	18 ± 0.4
Fe(III) Complex	19 ± 0.5	18 ± 0.4	18 ± 0.5	19 ± 0.4
Ciprofloxacin (Control)	27 ± 0.4	28 ± 0.5	27 ± 0.4	28 ± 0.5

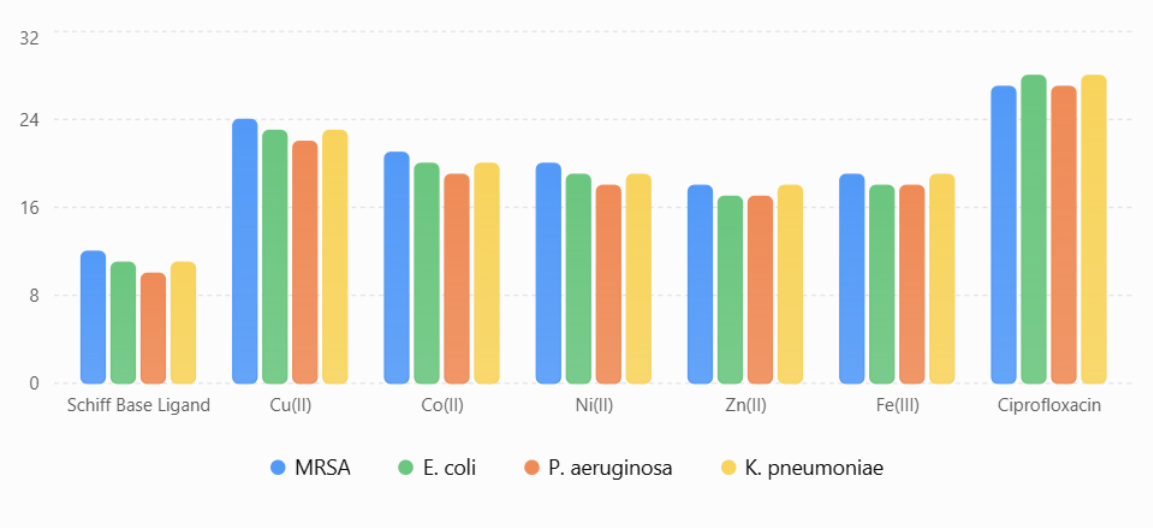

Figure 6. Comparative Antibacterial Activity of Schiff Base Ligand and Transition Metal Complexes

Minimum Inhibitory Concentration (MIC)

The MIC values confirmed the agar diffusion results. The Cu(II) complex exhibited the lowest MIC values against all tested microorganisms, indicating the strongest antibacterial potency. The Schiff base ligand showed comparatively higher MIC values, suggesting lower antimicrobial effectiveness.

Table 6. Minimum Inhibitory Concentration (MIC) of Synthesized Compounds (µg/mL)

Compound	MRSA	*E. coli*	*P. aeruginosa*	*K. pneumoniae*
Schiff Base Ligand	125	125	250	125

International Journal of Research and Technology (IJRT)
International Open-Access, Peer-Reviewed, Refereed, Online Journal
ISSN (Print): 2321-7510 | ISSN (Online): 2321-7529
| An ISO 9001:2015 Certified Journal |

Compound	MRSA	*E. coli*	*P. aeruginosa*	*K. pneumoniae*
Cu(II) Complex	15.6	15.6	31.2	15.6
Co(II) Complex	31.2	31.2	62.5	31.2
Ni(II) Complex	31.2	62.5	62.5	31.2
Zn(II) Complex	62.5	62.5	125	62.5
Fe(III) Complex	31.2	62.5	62.5	31.2

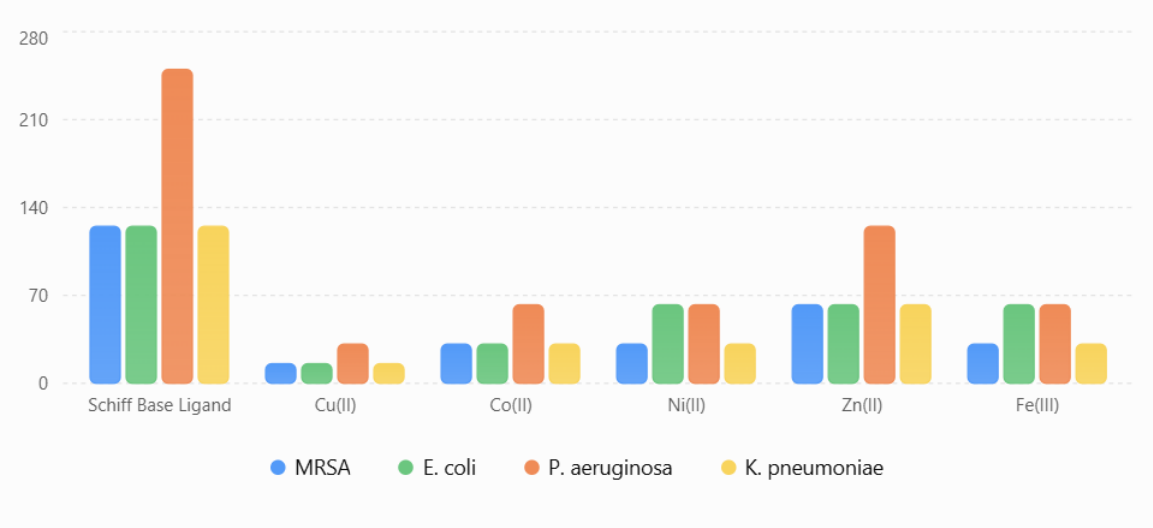

Figure 7. Minimum Inhibitory Concentration (MIC) of Schiff Base Ligand and Metal Complexes

The antibacterial results indicate that coordination of the Schiff base ligand with transition metal ions markedly enhanced antimicrobial activity. The Cu(II) complex exhibited the highest inhibition zones and the lowest MIC values, demonstrating superior effectiveness against all MDR bacterial strains. This enhanced activity is consistent with chelation theory, which states that metal coordination increases lipophilicity, facilitating penetration through microbial cell membranes and improving interaction with intracellular targets such as DNA and enzymes. Overall, the order of antibacterial activity observed was:

Cu(II) > Co(II) > Ni(II) > Fe(III) > Zn(II) > Schiff Base Ligand.

These findings suggest that transition metal–Schiff base complexes, particularly the Cu(II) complex, possess significant potential as antimicrobial agents against multidrug-resistant microorganisms.

4. DISCUSSION

The present investigation successfully demonstrated the synthesis, characterization, and biological evaluation of transition metal–Schiff base complexes with significant antimicrobial

potential against multidrug-resistant (MDR) microorganisms. The synthesized Schiff base ligand effectively coordinated with Cu(II), Co(II), Ni(II), Zn(II), and Fe(III) ions through the azomethine nitrogen and phenolic oxygen atoms, forming stable chelate complexes. The successful formation of these complexes was confirmed by FT-IR, UV–Visible spectroscopy, magnetic susceptibility, powder XRD, and SEM analyses. The observed spectral shifts, characteristic metal–ligand vibrations, and changes in morphology collectively verified the proposed coordination structures and enhanced stability of the synthesized compounds.

The antimicrobial evaluation revealed that transition metal coordination markedly enhanced the biological activity of the Schiff base ligand. Among all synthesized complexes, the Cu(II) complex exhibited the highest antibacterial activity, producing the largest zones of inhibition and the lowest MIC values against methicillin-resistant *Staphylococcus aureus* (MRSA), *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae*. The Co(II), Ni(II), Fe(III), and Zn(II) complexes also demonstrated improved antimicrobial efficacy compared with the free ligand, confirming that metal coordination plays a crucial role in enhancing biological performance.

The enhanced antimicrobial activity can be explained by the chelation theory, according to which coordination reduces the polarity of the metal ion while increasing the lipophilicity of the complex. Increased lipophilicity facilitates penetration through microbial cell membranes, allowing the complexes to interact more effectively with intracellular biomolecules such as DNA, proteins, and essential enzymes. Additionally, redox-active metal ions, particularly Cu(II), may promote the generation of reactive oxygen species (ROS), resulting in oxidative damage to microbial cells and inhibition of vital metabolic pathways. The ability of these complexes to target multiple cellular components may also reduce the probability of resistance development.

Overall, the findings indicate that transition metal–Schiff base complexes, especially the Cu(II) complex, possess promising antimicrobial properties against multidrug-resistant pathogens. These results support the potential application of metal-based Schiff base complexes as alternative antimicrobial agents and provide a strong foundation for future studies involving cytotoxicity assessment, molecular docking, in vivo evaluation, and the development of new metal-based therapeutic candidates for combating antimicrobial resistance.

5. CONCLUSION

The present study successfully demonstrated the design, synthesis, characterization, and biological evaluation of transition metal–Schiff base complexes as potential antimicrobial agents against multidrug-resistant (MDR) microorganisms. Spectroscopic and physicochemical analyses confirmed the successful coordination of the Schiff base ligand with Cu(II), Co(II), Ni(II), Zn(II), and Fe(III) ions, resulting in stable metal complexes with enhanced structural and thermal stability. Biological investigations revealed that all synthesized metal complexes exhibited superior antimicrobial activity compared with the free Schiff base ligand. Among the investigated compounds, the Cu(II) complex showed the highest antibacterial efficacy, producing the largest inhibition zones and the lowest minimum inhibitory concentration (MIC) values against the tested MDR bacterial strains. The enhanced biological

performance is primarily attributed to chelation, increased lipophilicity, and improved interaction of the metal complexes with microbial cellular targets. These findings highlight the significant role of transition metal coordination in improving the therapeutic potential of Schiff base ligands. Overall, the study demonstrates that transition metal–Schiff base complexes represent promising candidates for the development of next-generation antimicrobial agents. Further investigations involving molecular docking, cytotoxicity studies, pharmacokinetic evaluation, and in vivo experiments are recommended to establish their safety, efficacy, and potential clinical applications in combating antimicrobial resistance.

REFERENCES

1. LBoulechfar, C., Ferkous, H., Delimi, A., Djedouani, A., Kahlouche, A., Boublia, A., Darwish, A. S., Lemaoui, T., Verma, R., & Benguerba, Y. (2023). Schiff bases and their metal complexes: A review on the history, synthesis, and applications. *Inorganic Chemistry Communications, 150*, 110451. https://doi.org/10.1016/j.inoche.2023.110451
2. Kumar, R., Singh, A. A., Kumar, U., Jain, P., Sharma, A. K., Kant, C., & Faizi, M. S. H. (2023). Recent advances in synthesis of heterocyclic Schiff base transition metal complexes and their antimicrobial activities especially antibacterial and antifungal. *Journal of Molecular Structure, 1294*(Part 2), 136346. https://doi.org/10.1016/j.molstruc.2023.136346
3. Sonawane, H. R., Vibhute, B. T., Aghav, B. D., Deore, J. P., & Patil, S. K. (2023). Versatile applications of transition metal incorporating quinoline Schiff base metal complexes: An overview. *European Journal of Medicinal Chemistry, 258*, 115549. https://doi.org/10.1016/j.ejmech.2023.115549
4. Catalano, A. (2023). Schiff bases: A short survey on a promising scaffold in drug discovery. *Current Medicinal Chemistry, 30*(37), 4176–4180. https://doi.org/10.2174/0929867330666230201121432
5. Priya, B., Utreja, D., Sharma, S., Kaur, G., & Madhvi. (2023). Synthesis and antimicrobial activities of indole-based Schiff bases and their metal complexes: A review. *Current Medicinal Chemistry, 27*(11), 941–961. https://doi.org/10.2174/1385272827666230901140611
6. Panghal, M., Kumari, S., Goel, N., & Rani, I. (2023). Synthesis, spectroscopic characterization, antioxidant properties and antimicrobial activity of transition metal complexes from novel Schiff base. *Current Journal of Applied Science and Technology, 42*(5), 40–47. https://doi.org/10.9734/CJAST/2023/v42i54070
7. Madhukar, V., & Jyothi, S. (2023). Schiff base metal complex synthesis, characterization, and antimicrobial application. *Indian Journal of Chemistry, 62*(9), 976–979. https://doi.org/10.56042/ijc.v62i9.5380
8. Kumar, R., Seemab, K., Singh, D. K., Jain, P., & Manave, N. (2023). Synthesis, antibacterial and antifungal activities of Schiff base rare earth metal complexes: A review of recent work. *Journal of Coordination Chemistry, 76*(9–10), 695–724.
9. Sarwar, A., Bahron, H., Sherino, B., Ali, A., Bhangwar, S., & Alias, Y. (2023). A review of current trends of antibacterial Schiff base complexes: Lower and higher transition metal

complexes. *Malaysian Journal of Microbiology, 19*(3), 333–347. https://doi.org/10.21161/mjm.220103

10. Pervaiz, M., Sadiq, S., Sadiq, A., Younas, U., Ashraf, A., Saeed, Z., Zuber, M., & Adnan, A. (2021). Azo-Schiff base derivatives of transition metal complexes as antimicrobial agents. *Coordination Chemistry Reviews, 447*, 214128. https://doi.org/10.1016/j.ccr.2021.214128
11. Mukhtar, S., Hassan, A., Morsy, N., Hafez, T., Hassaneen, H., & Saleh, F. (2021). Overview on synthesis, reactions, applications, and biological activities of Schiff bases. *Egyptian Journal of Chemistry, 64*(11), 6541–6554. https://doi.org/10.21608/EJCHEM.2021.79736.3920
12. da Silva, C. M., da Silva, D. L., Modolo, L. V., Alves, R. B., de Resende, M. A., Martins, C. V. B., & de Fátima, Â. (2011). Schiff bases: A short review of their antimicrobial activities. *Journal of Advanced Research, 2*(1), 1–8. https://doi.org/10.1016/j.jare.2010.05.004