

**Biogenic AgNPs Decorated Cu(II) and Zn(II) Complexes of Imine-Quinoline
Ligand: Synthesis, Characterization and Biological Applications**



Gemechu Shumi Bekesho

**A Dissertation Submitted to
The Department of Applied Chemistry
College of Applied Natural Science**

**Presented in Partial Fulfillment of the Requirement for the Degree of
Doctor of Philosophy (PhD) in Applied Chemistry (Bioinorganic Chemistry)**

**Office of Graduate Studies
Adama Science and Technology University**

**November, 2024
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Main Supervisor: Prof. Tegene Desalegn (PhD)

Co- Supervisor: Prof. Taye Beyene (PhD)

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DECLARATION

I hereby declare that this PhD Dissertation entitled “**Biogenic AgNPs Decorated Cu(II) and Zn(II) Complexes of Imine-Quinoline Ligand: Synthesis, Characterization and Biological Applications**” is my original work under the supervision of **Professor Tegene Desalegn** and **Professor Taye Beyene**. It has not been presented for award of any a degree or diplomas in any other university, and all sources of material used for this dissertation have been duly acknowledged by means of completed reference.

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RECOMMENDATION

To: Applied Chemistry Department

Subject: Dissertation Submission

This is to certify that the PhD dissertation entitled “**Biogenic AgNPs Decorated Cu(II) and Zn(II) Complexes of Imine-Quinoline Ligand: Synthesis, Characterization and Biological Applications**” submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy in **Applied Chemistry (Bioinorganic Chemistry)**, the Graduate program of the Department of **Applied Chemistry**, and has been carried out by **Gemechu Shumi** under our supervision. Therefore, we recommend that the student has fulfilled the requirements and hence hereby he can submit the dissertation to the department for further review and defense.

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APPROVAL of PhD DISSERTATION

We hereby certify that the recommendation and suggestion given by the progress review committee are appropriately incorporated into the final PhD Dissertation entitled “**Biogenic AgNPs Decorated Cu(II) and Zn(II) complexes of imine-Quinoline Ligand: Synthesis, Characterization and Biological Applications**” by **Gemechu Shumi**

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LIST OF ACRONYMS AND ABBREVIATIONS

^{13}C NMR	Carbon Nuclear Magnetic Resonance
^1H NMR	Proton Nuclear Magnetic Resonance
AMR	Antimicrobial Resistance
ATCC	American Type Culture Collection
CADD	Computer-Aided Drug Design
CFU	Colony-Forming Unit
DDS	Drug Delivery System
DFT	Density Functional Theory
DMSO	Dimethylsulfoxide
DNA	Deoxyribonucleic Acid
DPPH	2, 2-diphenyl-1-picrylhydrazyl
EDX	Energy-Dispersive X-ray
ER α	<i>Estrogen Receptors Alpha</i>
FTIR	Fourier Transforms Infrared
FWHR	The Full Width Half Maximum
HOMO	Highest Unoccupied Molecular Orbitals
IARC	International Agency for Research on Cancer
IC50	Half Inhibition Concentration
IQL	Imine Quinoline Ligand
LALE	<i>Lippia Abyssinica</i> Leaf Extract
LUMO	Lowest Unoccupied Molecular Orbitals
MCF-7	Master Control Facility
MDR	Multi Drug Resistance
MHA	Muller-Hinton Agar
MHz	Mega Herz
MNPs	Metallic Nanoparticles
MTT	3-[4,5-dimethylthiazole-2-yl]-2,5-diphenyltetrazolium bromide
PDB	Protein Data Bank
pXRD	Powder X-ray diffraction

RNS	Reactive Nitrogen Species
ROS	Reactive Oxygen Species
SAS	Statistical Analysis System
SEM	Scanning Electron Microscopy
SOD	Superoxide Dismutase
SPR	Surface Plasmon Resonance
TEM	Transmission Electron Microscopy
TGA/DTA	Thermogravimetry Differential Thermal Analysis
UK	United Kingdom
US	United State
WHO	World Health Organization

ABSTRACT

Metal complexes have been the subject of extensive research in coordination chemistry since the discovery of metal-based medications. Furthermore, the combination of bioactive compounds with nanomaterial provides the opportunity in drug systems to overcome drug resistance, reduce drug-related side effects and develop new pharmaceutical agents. This dissertation aimed to synthesize and characterize of novel Cu(II) and Zn(II) complexes of imine derivative ligands of quinoline by condensing 7-chloro-2-hydroxyquinoline-3-carbaldehyde and 2,2'-thiodianiline, and decorated synthesized AgNP functionalized the amino acids with the metal complexes. The study evaluated the Cytotoxicity, antibacterial and antitoxic activity of the as-synthesized complexes. Furthermore, the molecular docking properties of these complexes were assessed. The synthesized pristine and silver nanoparticle decorated metal complexes were characterized using spectroscopic (UV-Vis, FTIR, ^1H and ^{13}C), microscopic (SEM-EDX and TEM), thermal (TGA/DTA) and diffraction (XRD) techniques. The biological activities of the synthesized ligand and its metal complexes, as well as their AgNP decorated compounds were evaluated in vitro for antibacterial activity against gram-positive (*Staphylococcus aureus*) and gram-negative (*Escherichia coli* and *Pseudomonas aeruginosa*) bacterial strains and for antioxidant activity using DPPH. In vitro anticancer activity against the MCF-7 human breast cancer cell line was examined for Cu(II) complexes. The molar conductivity and chloride test confirmed the non-electrolytic nature of the Cu(II) and Zn(II) complexes, supporting the proposed octahedral geometry. Furthermore, excellent correlation was observed between the experimental absorption peaks and those calculated using TD-DFT, validating the electronic structure predictions. The pXRD diffractogram analysis revealed that the synthesized ligand and its complexes were polycrystalline systems, with average crystallite sizes of nano level 13.28, 31.47, 11.57, 21.19, 16.26, 25.63, 20.67, 20.83, 19.74, 22.68, and 19.25 nm for imine quinoline ligand (L), CuL, and CuL₂, ZnL, ZnL₂, AgNP, His-AgNP@L, His-AgNP@CuL, His-AgNP@CuL₂, His-AgNP@ZnL, and His-AgNP@ZnL₂ respectively. The TEM micrograph revealed that the morphology of AgNPs had diameters ranging from 5 – 14 nm. The AgNP@CuL showed maximum growth inhibition of 12.00, 10.00, and 11.33 mm at 50 $\mu\text{g/mL}$ with the MIC of 12.5 $\mu\text{g/mL}$ against *S. aureus*, *E. coli*, and *P. aeruginosa*, respectively. Similarly, AgNP@CuL exhibited the highest antioxidant activities against DPPH radicals with a half inhibition concentration (IC_{50}) of $105.9 \pm 0.978 \mu\text{g/mL}$ stress. The CuL complex had the highest cytotoxic potency against MCF-7 breast cancer cells, with an IC_{50} of $43.82 \pm 2.351 \mu\text{g/mL}$. All the complexes had strong binding affinities according to the molecular docking investigation with L (-11.31 kcal/mol), ZnL (-11.38 kcal/mol), and ZnL₂ (-13.51 kcal/mol) against the active sites of *S. aureus*, and (-7.92, -8.07, and -8.92 kcal/mol, respectively) against *E. coli*, highlighting the Zn(II) complexes as potent in biological activity. Similarly the synthesized L and Cu(II) complexes (CuL and CuL₂) demonstrated significant binding interactions with *S. aureus*, *E. coli*, and *ER α* . CuL showing the strongest binding energy (-13.82 kcal/mol) and inhibition constant ($7.5 \times 10^{-5} \mu\text{M}$) against *S. aureus*, while CuL₂ exhibited a strong binding energy (-8.66 kcal/mol) and inhibition constant (0.45 μM) against *ER α* , scoring their potential as effective inhibitors.

Key words: Biogenic; decorated AgNP-metal complex; imine quinoline ligand; molecular docking; anticancer activity

CHAPTER ONE

1. INTRODUCTION

1.1. Background of the study

In the present era, the world faces a dual burden of communicable and non-communicable diseases, affecting both developed and developing countries (Boutayeb, 2010; Hambleton *et al.*, 2023). Globally, cancer incidence is expected to rise from 19.9 million new cases in 2022 to 21.3 million in 2025, reflecting a growing burden on society (Ferlay *et al.*, 2024; Siegel *et al.*, 2023). By 2040, cancer cases may exceed 28 million worldwide (Arnold *et al.*, 2022), with breast cancer being the most common diagnosis and leading cause of death among women (Sung *et al.*, 2021). Current cancer treatments, including platinum-based drugs like cisplatin, are often associated with severe side effects and drug resistance, leading to treatment failure (Lucaciu *et al.*, 2022; Zhang *et al.*, 2022). Similarly, drug resistance in treating infections has become a global challenge, contributing to rising morbidity and mortality (WHO, 2019; Evans & Kavanagh, 2021), and adding to the strain on global healthcare systems (Dhanda *et al.*, 2023; Muteeb *et al.*, 2023). Also, diseases such as cancer and infections are linked to oxidative stress, where an overproduction of reactive oxygen species (ROS) disrupts cellular redox balance, leading to damage of DNA, proteins, and membranes, and contributing to chronic illnesses and aging (Jomova *et al.*, 2023; Reddy, 2023). Furthermore, antimicrobial resistance can negatively impact cancer treatment and increase cancer-related mortality (Danielsen *et al.*, 2023). Given the rise of multidrug-resistant bacterial strains, researchers and pharmaceutical companies are focusing on developing effective treatments for infectious diseases (Dhanda *et al.*, 2023; Muteeb *et al.*, 2023). This highlights the urgent need for new anticancer and antimicrobial drugs with novel mechanisms of action that can overcome drug resistance, reduce side effects, and improve patient outcomes (ChunYan *et al.*, 2022). One promising avenue is the development of metallo-drugs that leverage the unique reactivity and redox chemistry of metal-based coordination complexes (Lucaciu *et al.*, 2022; Zhang *et al.*, 2022).

The coordination chemistry of transition metal complexes has significantly advanced biochemical sciences, particularly in healthcare (Venkatesh *et al.*, 2024). Since the discovery of cisplatin for cancer treatment, metal complexes have gained attention for their therapeutic potential, motivating the development of new metal-based drugs (Abd El-Lateef *et al.*, 2023; Hassan *et al.*, 2022). These complexes offer diverse molecular structures, coordination geometries, and kinetic properties that are not achievable with conventional carbon-based compounds (Cohen, 2007; Kostova, 2023). Their versatility, including variable oxidation states and ligand interactions, makes them attractive for therapeutic research (Jagessar, 2023). Metal-ligand interactions can be tuned to enhance drug targeting and efficacy, improving the absorption, accumulation, and direction of bioactive molecules (Boros *et al.*, 2020; Geisler *et al.*, 2021; Li *et al.*, 2021). These interactions, determined by metal ions' physical and chemical properties, play a critical role in drug design by influencing biological activity (Anthon *et al.*, 2020; Jurca *et al.*, 2017). Research is increasingly focusing on non-platinum metal complexes, such as those involving copper, zinc, and titanium, which offer lower side effects than platinum-based therapies and hold promise for chemotherapy and other treatments (Gamberi and Hanif, 2022; Todorov and Kostova, 2023).

As copper ions are important in cell division and growth and accumulated research evidence supports its potential utility in the battle against cancer (Guan *et al.*, 2023). Copper-based complexes have been also studied for their direct free radical scavenging abilities, demonstrating their potential to mitigate the harmful effects of oxidative stress (Hong *et al.*, 2024; Liu *et al.*, 2023). Copper can also enhance the effectiveness of some chemotherapeutic drugs like bioactive organic ligands by sensitizing cancer cells. This synergistic effect can improve the overall efficacy of cancer therapy (Aishajiang *et al.*, 2023). Similarly, over 3000 proteins, including transcription factors and enzymes, have been shown to include zinc as a structural, catalytic, and regulatory element. Zinc's function in intracellular and intercellular communication as a signaling and regulatory component has also come to light recently. Zinc ions are linked to 10% of the human proteome and are essential for controlling gene expression, DNA metabolism, chromatin structure, cell proliferation, maturation, death, immunological responses, and antioxidant defenses (Costa *et al.*, 2023).

Imine-based ligands, like quinolines, have generated significant interest due to their fascinating chemical properties and potential biological uses, including antibacterial, and anticancer effects (Pinheiro *et al.*, 2023; Goto *et al.*, 2019). Quinolines are versatile heterocyclic aromatic compounds containing nitrogen and various moieties (Marella *et al.* 2013). They are present in both synthetic and naturally occurring biologically active substances, making them crucial in pharmaceutical research for development of novel drugs (Kaur and Kumar 2021). Since quinolines have great potential to function as a bioactive reagents on their own, their transition metal complexes can further trigger increase their biological activity (Goto *et al.*, 2019; Ajani *et al.*, 2022). Quinoline motif Schiff bases, such as (*E*)-N-(furan-2-yl methylene) quinoline-8-amine and their metal complexes with Mn(II), Co(II), Ni(II), Cu(II), and Zn(II), have been reported to possess antimicrobial, antioxidant, and *in vitro* cytotoxicity activities (Shakir *et al.*, 2015). The significant number of studies carried on the structure-function relationship of metal complexes indicate that altering the ligand and the general structure of the complex significantly increases potency and efficiency of the metal complexes for various biological applications (Ibrahim *et al.*, 2024).

Many drugs and therapies are now available to treat fatal illnesses, but a major drawback is their lack of target specificity, often causing side effects in patients. Nanomaterial, particularly noble metal nanoparticles like AgNPs, offer-promising solutions for targeted drug delivery. Studies have shown that with minimal modifications, nanomaterial can be optimized to deliver drugs efficiently to specific cellular and intracellular targets. Due to their small size, nanoparticles can easily penetrate host cells and circulate throughout the body, making them ideal for delivering high doses of chemotherapeutic agents or therapeutic genes to precise locations. The surface stability of AgNPs further enhances their biomedical potential, allowing for surface modifications with biological and organic molecules to improve their functionality and efficiency (Gaikwad *et al.*, 2019).

Nowadays, finding an effective formulation of drugs with nanoparticles to treat microbial infections has become a critical issue due to the emergence of multidrug-resistant bacterial strains. Nanoparticles can make a new way to treat many diseases, being able to improve stability, promoting controlled liberation or decreasing cytotoxicity of medicaments. A highly

promising and innovative approach to deal with the bioavailability issue and to achieve even more diverse potential health benefits is the importance of nanoparticle-decorated ligands and its derivative metal complexes (David and Moldovan, 2020; Pillai *et al.*, 2020). The most commonly used agents for the surface functionalization of metal nanoparticles are amino acids (Ahmad *et al.*, 2022; Chakraborty *et al.*, 2018). The surface functionalization provides additional sites, such as amino or carboxyl groups, to act as a coupling agent between the nanoparticles and biomolecules. This enhances the stability of the nanoparticles and improves their activities. However, the combined activity of metal complexes and nanoparticles against bacteria has not been assessed well and could be an interesting approach for utilizing lower AgNPs concentrations for antibacterial activity.

Developing metal complexes decorated with functionalized metallic nanoparticles is vital for enhancing biological applications (Mondal *et al.*, 2024). Metal complexes, such as those based on Cu(II) and Zn(II), hold considerable therapeutic promise for treating conditions like infections and cancer. However, these complexes often face challenges related to stability, solubility, and selective targeting, which can limit their effectiveness. Integrating these metal complexes with functionalized AgNPs can address these issues by improving stability, solubility, and bioavailability. Functionalized AgNPs provide a stable platform for the metal complexes and can be modified to enhance biocompatibility, thereby allowing for targeted delivery and controlled release of the therapeutic agents (Wani *et al.*, 2016). This hybrid approach leverages the strengths of both components, offering a promising strategy to overcome the inherent limitations of metal complexes. By using functionalized AgNPs, the stability and efficacy of metal complexes are significantly enhanced, enabling more precise and effective treatment. The ability of AgNPs to improve targeted delivery and controlled release ensures that therapeutic agents are concentrated at the desired site, reducing off-target effects and toxicity. This combined system has the potential to advance treatment options for complex diseases, making it a valuable area for future research and development in therapeutic applications (Mondal *et al.*, 2024).

Moreover, the synergistic effect between the metal complexes and the functionalized AgNPs can lead to enhanced biological activity while minimizing the toxicity of silver nanoparticles.

The composite formation with imine ligands and Cu(II) or Zn(II) complexes allows for the use of a minimal amount of AgNPs, reducing the overall silver content, which in turn lowers the associated toxicity concerns. This controlled incorporation of AgNPs, combined with the therapeutic properties of the metal complexes, creates a highly effective system for biological applications, where the therapeutic benefits are maximized, and potential toxicity is minimized. Thus, the combination of metal complexes with functionalized AgNPs offers a novel, safer, and more efficient approach for targeted biological applications (Hajfathalian *et al.*, 2024; Yaqoob *et al.*, 2020).

1.2. Statement of the problem

Cancer remains one of the leading causes of mortality worldwide, with millions of new cases diagnosed annually. Despite advancements in cancer therapeutics, the effectiveness of current treatment strategies is severely limited by several challenges, including drug resistance, severe systemic toxicity, lack of selectivity for cancer cells, and diminished therapeutic efficacy. Additionally, oxidative stress, characterized by an imbalance between reactive oxygen species (ROS) and antioxidant defenses, is a major factor contributing to cancer progression, as well as various chronic and degenerative diseases. Conventional antioxidants, though widely used, suffer from issues such as low stability, limited bioavailability, and potential adverse effects at higher doses, highlighting the urgent need for novel and more effective therapeutic agents (Piña-Sánchez *et al.*, 2021; Anand *et al.*, 2023).

Additionally, The rate at which drug-resistant bacterial diseases are emerging and evading treatments is accelerating at an alarming pace, rendering it challenging the scientific community to keep pace with the rapid evolution of these pathogens. Several studies revealed that over 3 million people deceased from bacterial illnesses every year (Salam *et al.*, 2023). By 2050, drug resistant bacterial diseases are expected to kill 10 million people yearly, making them more dangerous than cancer (Tang *et al.*, 2023). Despite this growing risk, few antibiotics with limited mode of action are being produced, leading to a lack of drug to treat multidrug resistance (MDR) bacterial infections. Thus, the growing concern regarding drug-resistant bacterial strains and biofilm-associated infections, calls for the development of

advanced antibiotics. The WHO has called on nations to develop national strategies plan on how to minimize the development of antimicrobial resistance through the development of new novel antibiotics holding variable mechanism of action (Murray *et al.*, 2022; Nurul *et al.*, 2023). In addition, the problems of microbial infections, side effect of the drugs and the emergence of drug-resistant microbes are being responsible for the loss of countless number of lives and economic loss (Murray *et al.*, 2022; Nurul *et al.*, 2023).

Similarly, most diseases are linked ROS to as metabolic disorders such as cancer and other non-communicable diseases that antioxidants are the molecules that scavenge free radicals to combat cellular oxidative damage, which can slow or prevent the oxidation processes initiated by ROS that lead to carcinogenicity. However, the development of novel antioxidants with high pharmacological activity and few side effects remains challenging. Therefore, vast research on pharmaceuticals is necessary to develop a new therapeutic approach to discover promising and effective antibiotic drugs possessing novel modes of biological activities. Nowadays, the use of bioactive ligand like imine quinoline for the development of novel antioxidants, and antibacterial agents has caught the attention of researchers. This is because of the variety of their pharmacological activities including antioxidant, anticancer, and antimicrobial, as well as their ability to regulate the activity of several enzyme systems that participate in cell activation processes (Chaudhary *et al.*, 2023).

An imine quinolines derivative has pharmaceutical activity, involving direct scavenger of reactive oxygen species (ROS) and antimicrobial activities. Additionally, transition metal complexes with ligands containing both hard and soft donor groups have been used extensively to cause different types of damages to microbial cells because of membrane degradation, protein dysfunction and oxidative stress. These unique modes of action, combined with the wide range of geometries that metal complexes can adopt, make them suitable for the development of new antioxidant and antimicrobial drugs. Moreover, metal nanoparticles have synergy effect on antimicrobials that may target, facilitates transport, and increase its affectivities of the drug (Elebiju *et al.*, 2023; Hernández *et al.*, 2020). The review of literature revealed that there is no research being conducted on our title to develop new imine-quinoline ligand metal complexes and decorated with AgNPs for biological activity.

Thus, continuous research is desperately needed in order to come up with compounds which can be developed into new, cheap, less toxic and more efficacious antibacterial, antioxidant and anticancer drugs for compacting these diseases.

Therefore, this study addresses drug resistance challenges by synthesizing imine quinoline derivative ligand and its Cu(II) and Zn(II) complexes, and further enhancing their functionality through decoration with AgNPs. By using plant extracts for AgNP synthesis, functionalizing these nanoparticles with amino acids, and integrating them with the ligands and their metal complexes, the study aims to enhance antimicrobial efficacy while reducing toxicity. This approach tackles drug resistance by improving the stability and bioactivity of the metal complexes and creating synergistic effects that boost the therapeutic potential of the combined components. The research advances the application of coordination compounds and nanomaterials by demonstrating how tailored metal complexes and AgNPs can be optimized for better performance and safety, offering more effective and safer solutions in biological application.

1.3. Objective of the study

1.3.1. General objective

The main objective of this research work was to synthesize, characterize and evaluate the biological activities Cu(II) and Zn(II) metals complexes of imine derivative ligands of quinoline and decorated with biogenic silver nanoparticles (AgNPs).

1.3.2. Specific objectives

The specific objectives of this study were to:

- ❖ Synthesize the imine quinoline derivative ligand from 7-chloro-2-hydroxyquinoline-3-carbaldehyde and 2,2'-thiodianiline.
- ❖ Synthesize silver nanoparticles functionalized with phenylalanine and histidine using *Lippia Abyssinica* (Koseret) leaf extract.

- ❖ Synthesize the copper (II) and zinc (II) complexes with the synthesized ligand of imine quinoline derivative.
- ❖ Synthesize the ligand and its copper (II) and zinc (II) complexes decorated with biogenic AgNP.
- ❖ Characterize the synthesized compounds using FT-IR, UV–Vis, XRD, SEM-EDX and TEM, HR-TEM, NMR, TGA/DTA and TD-DFT.
- ❖ Evaluate *in vitro* antioxidant activities using *DPPH* and antibacterial activities using disk diffusion method for the synthesized ligand, its Cu(II) and Zn(II) complexes, and AgNP and metal complexes decorated with AgNP.
- ❖ Evaluate *in vitro* anticancer activities of synthesized ligand and its Cu(II) complexes against MCF-7 cell lines using MTT assay.
- ❖ To compute the molecular docking activities against *Staphylococcus aureus*, *Escherichia coli* and *Estrogen receptor alpha (ERα)* residual amino acids for the synthesized ligand and its Cu(II) and Zn(II) complexes.

1.4. Significant of the study

The findings of this study is necessary to provide basic understanding of the metal complexes and complexes decorated with AgNPs efficacy and disseminate the widespread uses in health care systems. This study lays foundation for future investigation into the field of pharmacy and other clinical actions for drugs discovery. Besides, this study was also expected add knowledge about the synthesis of imine quinoline derivative, its metal complexes biogenic of AgNPs and the metal complexes decorated with the biogenic AgNPs, which are ecofriendly.

1.5. Beneficiaries

This study helps researchers to get reliable information on bioactive imine-quinoline derivative ligand, and their metal complexes as a baseline for further investigations. In addition, research institutions academia, pharmaceuticals, local communities that are working on bioactive ligand and their metal complexes for medications could use the data to strengthen their believe in using imine-quinoline derivative ligand as a source for medications, and

researchers interested in coordination chemistry and their biological applications and related studies could be the primary beneficiaries of this study.

1.6. Outcome of the study

Currently, researchers gave great attention to microbial-causing infectious diseases for the reason of the increase in drug-resistant and newly developed pathogens spreading throughout the world. Even if there are some commercial antibiotic drugs, it is not easy to overcome drug-resistant microbial. Thus, this study was planned to synthesize Cu(II) and Zn(II) complexes with imine quinoline derivative ligand including AgNP decoration and evaluate their performance for biological application with a focus to produce enhanced antimicrobial, antioxidant and cytotoxic agents. The outcome shows that the targeted metal complexes of imine-quinoline derivative ligand derivatives were effective against selected gram negative and gram-positive bacterial strains as well as radical scavenging and cytotoxic activity. Furthermore, this study confirmed that there is a potential in the design and synthesis of metal complexes of imine quinoline ligand decorated with biogenic AgNP drug development. The results of the study were also published in three internationally reputable Scopus, and web of science-indexed journals as a platform to communicate the findings to the scientific community.

CHAPTER TWO

2. LITERATURE REVIEW

2.1. Antibacterial and Anticancer agents

2.1.1. Antibacterial drugs

Antimicrobial resistance is defined as the ability of microorganisms to counteract the action of antimicrobial agents and this phenomenon occurs when an antibiotic loses its efficiency to inhibit the bacterial growth (Prestinaci *et al.*, 2015; Uddin *et al.*, 2021). Unluckily, the rate of development of new antibiotics is lagging behind the rate of emergence of antibiotics resistance while, cost-effective global access to antibiotics is another challenge for health management systems in developing countries. Antimicrobial resistance impends the fundamental of modern medicine to the continuing hazard from infectious diseases. In fact, advancements in modern medicine, such as many medical and surgical procedures have facilitated by developing and using clinical classes of antibiotics (Coque *et al.*, 2023; Muteeb *et al.*, 2023).

Nowadays, the antibiotic era has threatened due to the alarming rate of multidrug resistance (MDR). Furthermore, secondary infections by MDR in complicated conditions like cancers may increase the mortality. Hence, to address the persistent global issues associated with MDR-related infectious diseases, there is pressing needs to develop new antimicrobial agents (Ahmed *et al.*, 2024; Kumar *et al.*, 2024).

Treatment of infection diseases caused by microbial remains an issue and challenging problem due to the combined effects of factors such as emerging infectious diseases and the growing number of multi-drug resistant microbial pathogens. Microbial infections are one of the leading diseases that are caused millions of deaths reported every year because of the lack of effective antimicrobial therapy due to microbial resistance towards conventional antibiotics (Murray *et al.*, 2022; Nwobodo *et al.*, 2022).

The quinolone and their imine base derivatives are an important class of organic ligands, which can be obtained naturally as well as synthesized from different sources of other organic molecules such as aldehyde and ketone groups possessing a wide range of biological activities (Al-Ostoot *et al.*, 2021; Dine *et al.*, 2023). According to Gemechu *et al.*, (2022) issues of antimicrobial resistance against commonly used antibiotics are becoming critical and a worldwide problem. Considering the high rising of drug-resistant microbial, developing another strategy to fight against bacterial infections. Therefore, searching for a novel and an effective antimicrobial agent is important to overcome this problem. Accordingly, metal complexes of imine quinoline derived organic ligands; decorated AgNPs with the metal complexes that metal complexes and their AgNPs decorated showed good antibacterial activity against *Escherichia coli*, *Pseudomonas aeruginosa*, and *Staphylococcus pyogenes*.

Furthermore, chelation may enhance or raise the biochemical potential of bioactive organic molecules (El-Lateef *et al.*, 2023; Sorrenti *et al.*, 2023). The character of the metal ion coordinated to the organic ligand may have its role in increasing the lipophilic character of the central metal ion, because of the partial sharing of its positive charge with the donor groups and possible π -electron delocalization over the chelate ring (Abate *et al.*, 2022; Saiyed *et al.*, 2023). This makes the permeation of metal complexes compounds through the lipid layer of the cell membrane easier. The increase in lipophilicity of the ligand up on metal coordination may contribute to its simplistic transport into the bacterial cell which blocks the metal binding sites in the enzyme of microorganisms (Claudel *et al.*, 2020; Holden *et al.*, 2021).

2.1.2. Anticancer agents

Cancer is a group of diseases in which the constant proliferation of neoplastic cells results in tumor formation. These tumors can invade the surrounding tissues and spread to distant organs. There are more than 200 different types of cancer and according to the World Health Organization (WHO), cancer is a significant cause of morbidity and mortality worldwide, regardless of the level of human development. By the end of 2020, there were 19.3 million new cancer cases reported and around 10.0 million cancer-related deaths. Hanahan and

Weinberg, in *The Hallmarks of Cancer and Hallmarks: The Next Generation of Cancer* emphasized that cancer is a complex, heterogeneous, and multifactorial disease. Recognizing this complexity provides critical insights into molecular mechanisms, guiding the development of more effective and targeted therapies. Most of the drugs against cancer are directed against a limited number of well-established targets (proliferation or cellular death), which reflects the difficulty, time, and cost involved in identifying and validating new therapeutic targets. Additionally, it is important to consider that cancer progression toward aggressive pathological stages or worse prognosis involves invasion and metastasis processes (Gonzalez-Ballesteros *et al.*, 2022; López-Otín *et al.*, 2023; Malik *et al.*, 2022; Moses *et al.*, 2018; Werner and LeRoith, 2022; Weth *et al.*, 2024; Yang *et al.*, 2022).

Cancer is a more complicated and fatal disease that poses a threat to human health all over the world. It involves multiple genes and crosstalk between signaling networks, changes in DNA, and/ or gene transcription and/or translation (Cao *et al.*, 2024). Cancer is caused by the malfunctioning of normal cell genes, which promotes uncontrolled proliferation, metastasis, and the rapid expansion of aberrant cells that grow beyond their normal limits. In the year 2020, the US was anticipated to witness 606,520 deaths and 1.8 million new cases. About 4.5 million premature cancer-caused deaths were reported worldwide in 2016 (American Cancer Society, 2020). Based on current data, the International Agency for Research on Cancer (IARC) forecasts that around 13 million cancer-related deaths would occur by 2030, prompting researchers to continue their hunt for an effective anticancer medicine. The most common clinical approaches for cancer treatment are chemotherapy, radiation therapy, surgery, hormone therapy, and targeted therapy with anticancer drugs. All cancer therapies have the potential to harm normal cells while destroying cancer cells. Targeted therapy, on the other hand, is a potential choice since normal cells can survive when cancer cell proliferation is reduced during targeted therapy. The damaged proteins produced by these mutated genes could be treated with targeted therapy (Araghi *et al.*, 2023; Waarts *et al.*, 2022; Wang *et al.*, 2023).

2.2. Metallo-drugs

Metal and metal-based antimicrobial substances have great potential against antimicrobial resistance pathogens. Metals are reported to target multiple cellular sites such as cellular membrane, genetic material, and reactive oxygen species-mediated cellular pleiotropic effects on microbial cells, contrary to organic antibiotics that act on specific targets on biochemical pathways such as replication, transcription, translation, and enzymatic reaction. Hence, there is a need for the development of novel, wide spectrum antimicrobial agents that could target and eliminate antibiotics resistance microbes. A renewed interest in metals as antimicrobial and biocide agents is reflected in hopes that less resistance will evolve (Gonzalez-Ballesteros *et al.*, 2022; Sharma *et al.*, 2022; Turner, 2017; Yang *et al.*, 2022).

Different metal complexes have their respective biological roles, and therefore, their design may help in developing new diagnostic probes as well as medicines. Metal complexes have emerged as great alternatives to organic compounds as they have specific steric and electronic effects that lead to different mechanisms of action (e.g., electron transfer and redox processes) (Boros *et al.*, 2020). Metals, being less electronegative, tend to form positively charged ions promptly, and this property lends them greater solubility in biological fluids. The positively charged ions thus formed have affinity for electron-rich biomolecules, such as DNA and proteins, and play an important role in stabilizing/influencing their tertiary or quaternary structures (Fonseca *et al.*, 2024; Pucelik *et al.*, 2024).

Metal ions interact with cancer cells through mechanisms involving DNA, proteins, membranes, and redox processes (Figure 1). They bind DNA at nucleobase sites like guanine's N7, disrupting replication and transcription, often inducing apoptosis through DNA crosslinking or strand breaks. Metal ions also target proteins by coordinating with thiols, histidine residues, or metal-binding motifs, affecting enzymes and signaling pathways such as PI3K/AKT, while disrupting redox homeostasis by inhibiting antioxidant enzymes like superoxide dismutase (SOD). At cellular membranes, metal ions interact with phospholipid head groups and membrane proteins, altering fluidity and inducing ROS production, which damages lipids, proteins, and DNA. Additionally, metals exploit cancer-specific biomarkers,

such as transferrin receptors, to selectively enter cells and trigger localized oxidative stress or apoptosis. Chelation mechanisms further disrupt essential metal-dependent processes, inhibiting angiogenesis and energy metabolism. These interactions underlie the efficacy of metal-based drugs like cisplatin, copper, and zinc complexes in targeting cancer pathways (Dasari and Bernard, 2014; Singh *et al.*, 2019; Van Der Westhuizen *et al.*, 2021).

The production of metal complexes as medications or diagnostic agents has piqued the interest of medicinal chemistry. A metal complex, also known as a coordination compound, is made up of a core metal atom and an array of molecules or anions known as ligand. Metal compounds provide therapeutic action mechanisms that organic agents cannot achieve due to their vast range of coordination numbers, geometries, and their kinetic properties. Preclinical studies *in vitro* and *in vivo* have investigated various metal compounds as antitumor medicines. Complexes with ionic organic ligands, classical inorganic agents, and increasingly all organometallic species are present in the various agents examined. Main group metals (such as bismuth, tin, antimony), transition metals (such as rhodium, vanadium, iron, cobalt, and gold), and cerium are among the metals studied. In contrast to organic molecules, metals provide accessible redox states, a wide range of coordination numbers, and ligand substitution kinetics for the design of anticancer medicines (Karges *et al.*, 2022; Patil *et al.*, 2022; Sodhi and Paul, 2019; Tesauero, 2022).

Metal complexes have created new challenges in terms of pharmacological limitations including low water solubility and reduced bioavailability. To attain a greater therapeutic index, breakthrough metal complexes must boost the selectivity of tumor cells over healthy cells, in addition to improving solubility. Targeting ligands, which are coupled to metal and carry the medicine to the interesting cells, can provide precise delivery. A targeted delivery system is defined as the ability of a molecule (ligands or metal complex) to be recognized by membrane receptors on tumor cells, resulting in efficient accumulation into tumor tissue. This build-up can also be caused by tumor cells' high need for a substance that isn't or lesser required by normal cells, like vitamins and sugar (Gemechu *et al.*, 2022; Karges *et al.*, 2022; Patil *et al.*, 2022; Sun *et al.*, 2023).

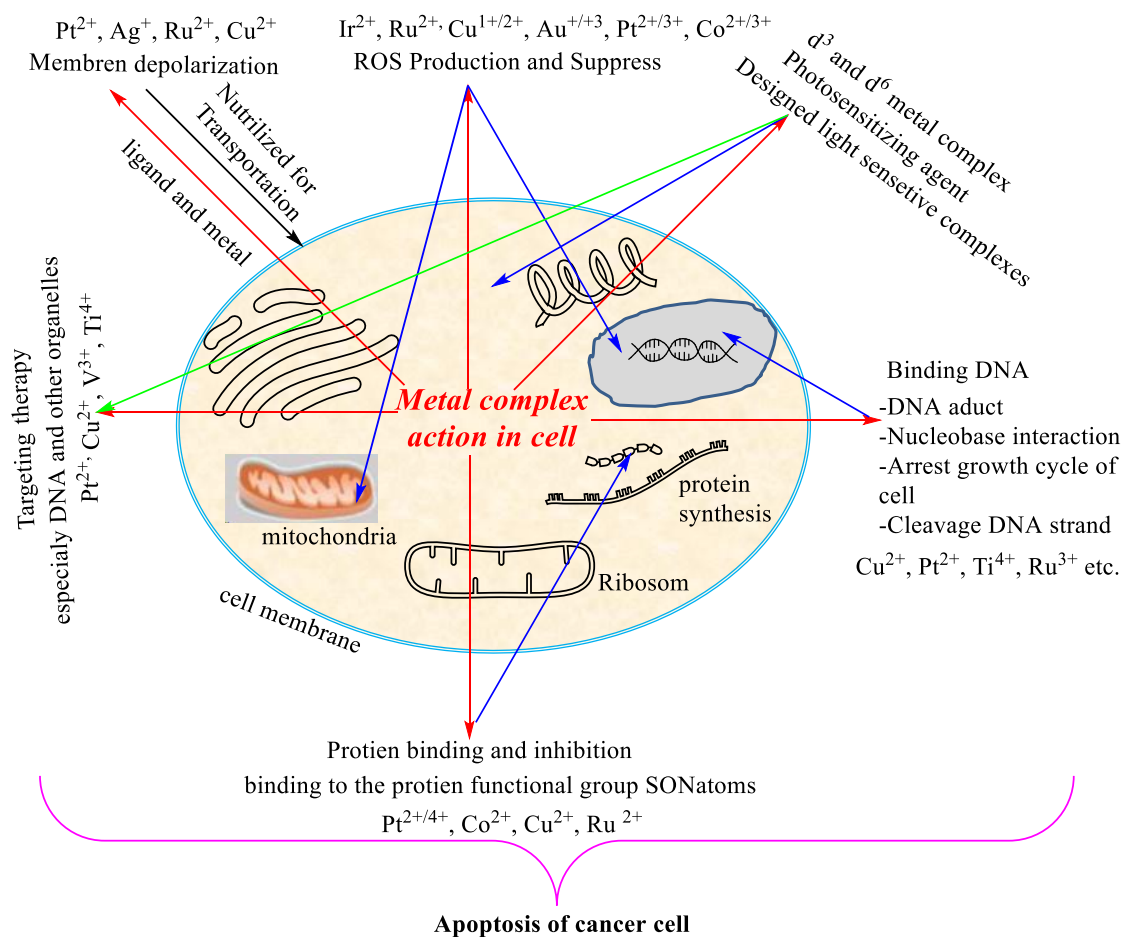


Figure 1. Mechanism of metal ion interactions sites in cancer cells

Metals have distinct properties such as various coordination modes, redox activity, and reactivity, and are required for a variety of metabolic activities in cells. Their concentration is tightly regulated inside the cells due to their reactivity. These metals could produce reactive oxygen species (ROS), which are important in cell metabolism, signaling for proliferation, differentiation, and cell death, and are a part of the cellular redox balance. Furthermore, ligand substitution and modification of existing chemical structures resulted in the synthesis of a wide spectrum of metal-based compounds, some of which have a better cytotoxic and pharmacokinetic profile than others. Figure 2 depicts the mechanism and specific function of metal complexation in increased drug-metal complex efficacy or potency (Smith *et al.*, 2023; Sun *et al.*, 2023; Timoshnikov *et al.*, 2022).

Many anticancer drug applications had been designed to apply as cancer cell reductase enzymes for target-specific activation. Some may also comprise metal complexes as pro-drugs for bio-reductive activation. For instance, cobalt complexes are reported to be bio-reductive pro-drugs, having a completely inert oxidized cobalt (III) state and really labile reduced cobalt (II) state. The reduction of Pt(IV) meta complex is an inert pro-drug and its reduction yields an active Pt(II) complex and equivalents of active ligand. Metal complex-drug conjugation can grow the potency and the performance of the drug. Curcumin is a bioactive ligand and has poor stability and bioavailability. However, the coordination of curcumin to an oxovanadium (IV) dipyrrophenazine (dppz) complex was found to prevent the hydrolytic cleavage of curcumin under physiological conditions and increases the efficiency of curcumin to target DNA cleavage. The metal complex can itself be biologically active. With the active metal complex and active organic molecules from a single prodrug, more than one way of acting upon a target can be realized. Synergy action drugs can be more potent than the parent organic drug and be able to avoid drug resistance mechanisms (Banik *et al.*, 2014; Chen and Hu, 2008; Mathuber *et al.*, 2020).

Conjugation of a drug to a positively charged metal complex has been shown to greatly enhance the aqueous solubility of the drug because of its hydrophilicity. Alternatively, conjugation of metal to negatively charged groups can reduce the negative charge of a drug, enhancing passive cellular uptake and absorption. Those are effective approaches to enhancing both the pharmacokinetic and pharmacodynamic properties of the parental drug and metal conjugated drugs to enter the cell. Reducing side effects and booming of the bioavailability of the metal-drug complexes can mainly depend on changes in environmental situations which include the redox status of the metals and the pH conditions (Figure 2) (Ndagi *et al.*, 2017).

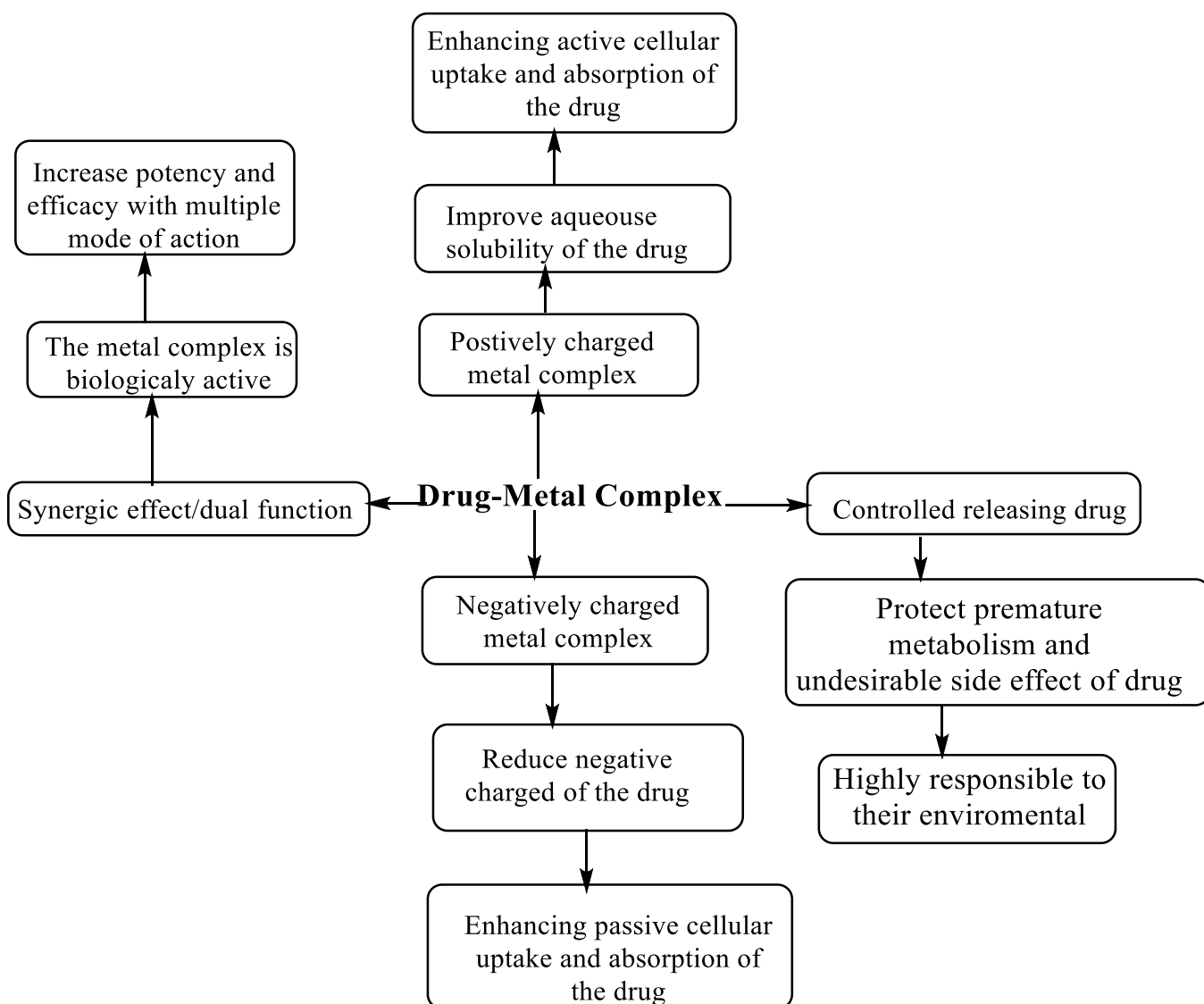


Figure 2. Illustration the mechanism and potency of drug metal complex

2.2.1. Copper and copper complexes

Copper is an essential component of a biological system, in which many chemicals need it to function properly. The coordination chemistry of copper has provoked the interest of numerous researchers due to its well-informed biological characteristics. There has been an interest in the medical uses of copper, as a complexing ion of known biologically active ligands and drugs. Some copper (II) complexes based on Schiff bases have been successfully synthesized and utilized as models in biological systems. They find many applications, mainly

as antimicrobial, antioxidant, catalytic, and anti-inflammatory agents. It has been reported that copper (II) complexes of thiosemicarbazone-based derivatives showed antimicrobial activities. The thiosemicarbazone derivative of copper (II) has good activity in the killing of *S. aureus*, *Salmonella Typhimurium*, and *Klebsiella pneumonia* after 6 h of incubation. Copper (II) complexes of semicarbazones and thiosemicarbazones with a different coordination mode through, O, N, and S donor atoms have been synthesized and evaluated for antimicrobial, antifungal, and antioxidant activities showing promising potential (Souza *et al.*, 2021).

Numerous studies have explored the antimicrobial, antioxidant, anti-inflammatory, and anticancer properties of copper (II) complexes. Antibacterial drugs commonly serve as ligands to form stable copper complexes. Notably, the antimicrobial activity of a copper-ciprofloxacin complex against *Mycobacterium smegmatis* bacteria was found to be three times higher than that of free ciprofloxacin, likely due to enhanced drug diffusion through cell membranes facilitated by the copper ion, possibly increasing the drug's lipophilicity. Additionally, the copper complex demonstrated potential against *Streptococcus aureus*, attributed to the intracellular reduction of Cu(II) to Cu(I), which activates toxic oxygen species and allows for the controlled release of ligands within the bacterial cell (Hussain *et al.*, 2019).

Moreover, copper (II) complexes of sulfacetamide, (N-[4-(aminofenil) sulfonyl] acetamide), sulfanilamide, and sulfisoxazole have been reported to show promising antibacterial, antifungal, and antioxidant activity results.*et al.* The copper (II) complexes of benzimidazoles have shown strong antibacterial activity against *Staphylococcus Epidermidis* and against fungi. The enhancement of antimalarial activity by copper coordination to ligands is commonly due to the reduction of Cu(II) to Cu(I) *et al.* Copper coordination of derivatives of salicylate has been one of the most often synthesized compounds in the search for biologically active compounds. Copper is an essential metal needed by organisms for many functions but can be toxic in large concentration. There are many copper-containing proteins present in microbes where copper acts as an electron donor/acceptor due to its ability to switch between copper (II) and copper (I) ions. To improve its antimicrobial activity, many researchers studied the coordination of organic molecules with copper. There are different mechanisms of action that depend on the geometry of the complexes and the nature of the ligand. Although the exact

mechanism of the antimicrobial activity of copper is not known, many investigations have shown that reactive oxygen species (ROS) produced through Fenton-type reactions damages DNA. The release of copper ions causes inactivation of enzymes that leads to its toxicity ref.

Simunkova and collaborators (2019) investigated the anticancer properties of copper (II) complexes containing flufenamic acid, mefenamic acid, tolfenamic acid and 1,10-phenanthroline (Figure 3). The complexes interact with albumin as targeting biomolecules, and the targeted drug delivery vehicles for Cu(II) complexes have a high affinity binding constant ($K = 106$). Once the albumin-bound complexes arrive at their destination, then the metal complex is freed from the carrier and bind to DNA. Cu(II) complexes are thus good superoxide dismutase (SOD) mimics as well as effective DNA intercalation. Furthermore, their ability to promote DNA damage is due to a redox-cycling mechanism that produces hydroxyl radicals, singlet oxygen, and superoxide radical anions (Ibrahim *et al.*, 2018; Simunkova *et al.*, 2019).

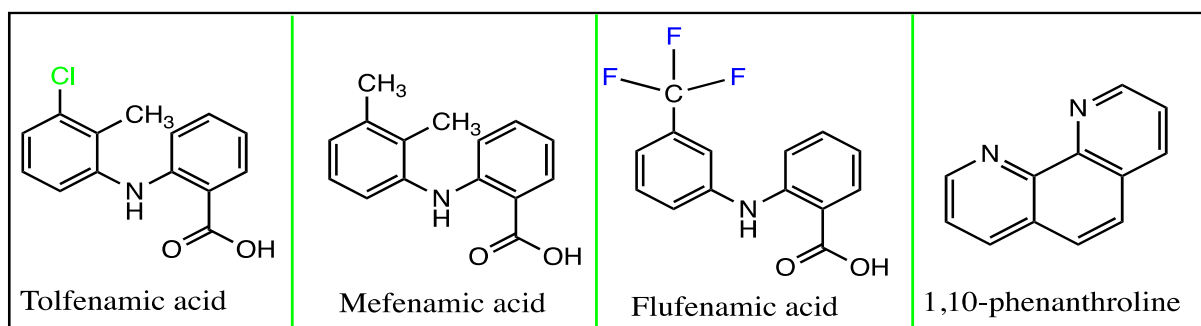


Figure 3. Structure of active anticancer fenamic acid derivatives and phenanthroline

2.2.2. Zinc and Zinc complexes

Zinc is one of the essential transition metals found throughout human body. It is the most abundant transition metal in the brain after iron. The importance of Zn^{2+} ions in metalloenzymes and metal-based medicines has been highlighted by numerous studies. Maintaining zinc's (Zn) ideal level in the human body is crucial for normal growth and development. Zinc is a highly essential trace element. The human body has zinc in the skin (5%), bones (30%), and muscles (60%). Among its many roles are its participation in the activation of several enzymes and proteins; moreover, it aids in the absorption of folate,

vitamin E, and A. Degenerative illnesses and infections may be more likely to occur in those with low zinc levels. The daily need for zinc varies based on an individual's sex and life stage, and like other micronutrients in the mineral group, it ranges from 2 to 13 mg (Caruso *et al.*, 2024).

Zinc is a common element in biological systems, mostly because of its unique physicochemical properties that set it apart from other first-row transition metals. With a d^{10} structure and diamagnetic, strong Lewis acid properties, the di-cationic redox inactive Zn^{2+} may sustain many coordination geometries and is prone to rapid ligand exchange. One of the spectroscopically silent metals in biology, zinc is considerably imitted in the scope of experimental methods relevant to its research in biological systems by its diamagnetism, lack of d-d transition, and white hue (Akbari *et al.*, 2024; Authors, 2024; Pellei *et al.*, 2021; Thompson, 2022).

Most of the Zn(II) complexes of semicarbazone and thiosemicarbazone ligands reported are showing tetrahedral as well as octahedral geometrical arrangements. The ligands are known to be bidentate or tridentate in their coordination mode. The ethyl acetoacetate semicarbazone and thiosemicarbazone are found to be tridentate in their Zn(II) complex in which the third coordinating center is provided by the acetyl carbonyl group. The discovery of Zn(II) complexes of thiosemicarbazones gave promising antimicrobial activities. The zinc (II) complexes of picolinaldehyde-S-methyl-and-S-benzylthiosemicarbazones, which were active in comparison to their metal-free ligands, were reported. Zinc (II) complex with the ligand (1H-imidazole-2-carboxaldehyde) thiosemicarbazone is recently reported as a candidate for antimicrobial and antioxidant agents (Akbari *et al.*, 2024; Fern *et al.*, 2023; Khalil *et al.*, 2014; Maret, 2016; Thompson, 2022).

Zinc (II) complexes of carboxylates were reported with good antibacterial activity; a strong inhibitive effect on *E. coli* and *S. aureus*. In the search for zinc (II) complexes of known antimicrobial drugs, strong antibacterial activity has been achieved. The various nitrogen-donor ligands have been studied and the complexes were found more potent than the drugs alone. In addition, zinc (II) complexes of salicylalimine were found to be active against *S.*

aureus. The zinc (II) complexes of sulfa-drugs, such as sulfadiazine and sulfamerazine also reported to be effective as antifungal agents against *Aspergillus* and *Candida* spp. The Zinc (II) complexes with amino acids have been also developed with antimicrobial activity. The zinc complex of pyrithione is a good example of an agent of antibacterial and antifungal activity. In the form of bis(N-oxopyridine-2-thionato) zinc (II) the drug inhibits the membrane transport processes by blocking the proton pump in fungi (Al-qadsy *et al.*, 2023; Chkirate *et al.* 2020; Dhabale *et al.*, 2022; Sovari and Zobi, 2020; Szunyogová *et al.*, 2007).

There are also reports of novel Zn(II) complexes of heterocyclic Schiff base ligands showing promising antimicrobial and antifungal agents. Turan and Sekerci, (2010) reported that the synthesis of octahedral Zn(II) complexes, Zn(II) chelates with the bidentate Schiff bases derived from 1,3,4-thiadiazole derivatives with salicylaldehyde and terephthalaldehyde having the coordination of protonated ethanol (Figure 4) (Behera *et al.* 2024; Turan and Sekerci 2010; Yamgar *et al.* 2014).

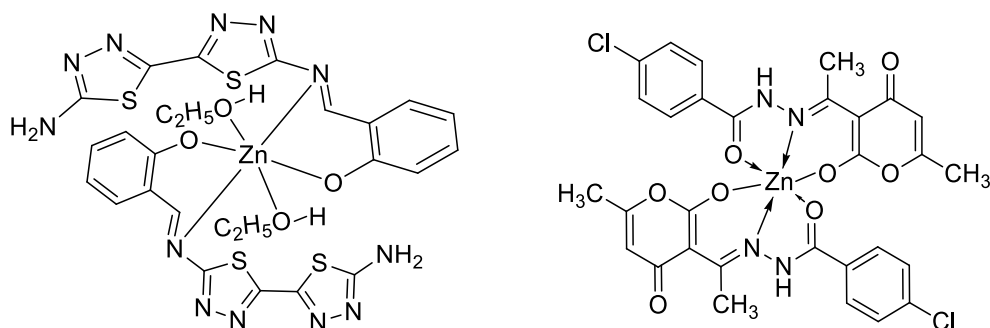


Figure 4. Zn(II) complexes with imine functional group containing ligands

2.3. Imine-Quinoline and its derivatives

Quinoline, benzene fused nitrogen heterocyclic system composed of two fused six-membered aromatic rings (benzene and pyridine) as shown Figure 5 (I), is found in many drug molecules, plays an important role in drug development, and has always remained as lead in the field of pharmaceutical prospective (Ayala *et al.*, 2023). Quinoline nitrogen heterocyclic acting as a weak tertiary base and could form salts with acids and undergoing electrophilic substitution reactions as well as reactions similar to those of pyridine and benzene. It is one of the most privileged N-containing motifs till date, most found in various natural products, most common

being Cinchona alkaloids and pharmacologically active substances thereby displaying a broad range of biological activity. Along with their rising significance in many other fields, they have also made major contributions to society through their usage as medicinal agents, applications in veterinary and human medicine, and molecular engineering. In addition, their exploration contributes to discovering and developing new drugs for various diseases, addressing unmet medical needs, and improving patient outcomes. The unique chemical structure of quinoline and the ability to modify its substituents have enabled researchers to design novel and potent quinoline-based drugs. The versatility of quinoline in exhibiting a multitude of pharmacological responses has captured the attention of medicinal chemists to explore its vast potential in various therapeutic applications (Bheemayya *et al.*, 2023).

In medicinal chemistry, understanding the structure-activity relationship (SAR) is critical for designing and developing new drugs with improved pharmacological activity. SAR studies involve evaluating the relationship between the chemical structure of a molecule and its pharmacological activity. Researchers have conducted numerous SAR studies of quinoline derivatives to identify the specific structural features that are responsible for their diverse pharmacological effects. In the case of quinoline-based drugs, SAR studies have identified the importance of the substitution pattern around the quinoline ring for anticancer and antibacterial activity that includes Brequinar (**II**) (anticancer), Ciprofloxacin (**III**) (antibacterial), antioxidant (**V** and **VI**) and Lenvatinib (**IV**) (anticancer) in (Figure 5) were reported. Overall, SAR studies have provided valuable insights into the molecular mechanisms underlying the pharmacological activity of quinoline derivatives. By identifying the specific structural features responsible for a compound's pharmacological activity, researchers can design and develop new drugs with improved efficacy and reduced side effects (Bheemayya *et al.*, 2023; Hamama *et al.*, 2018).

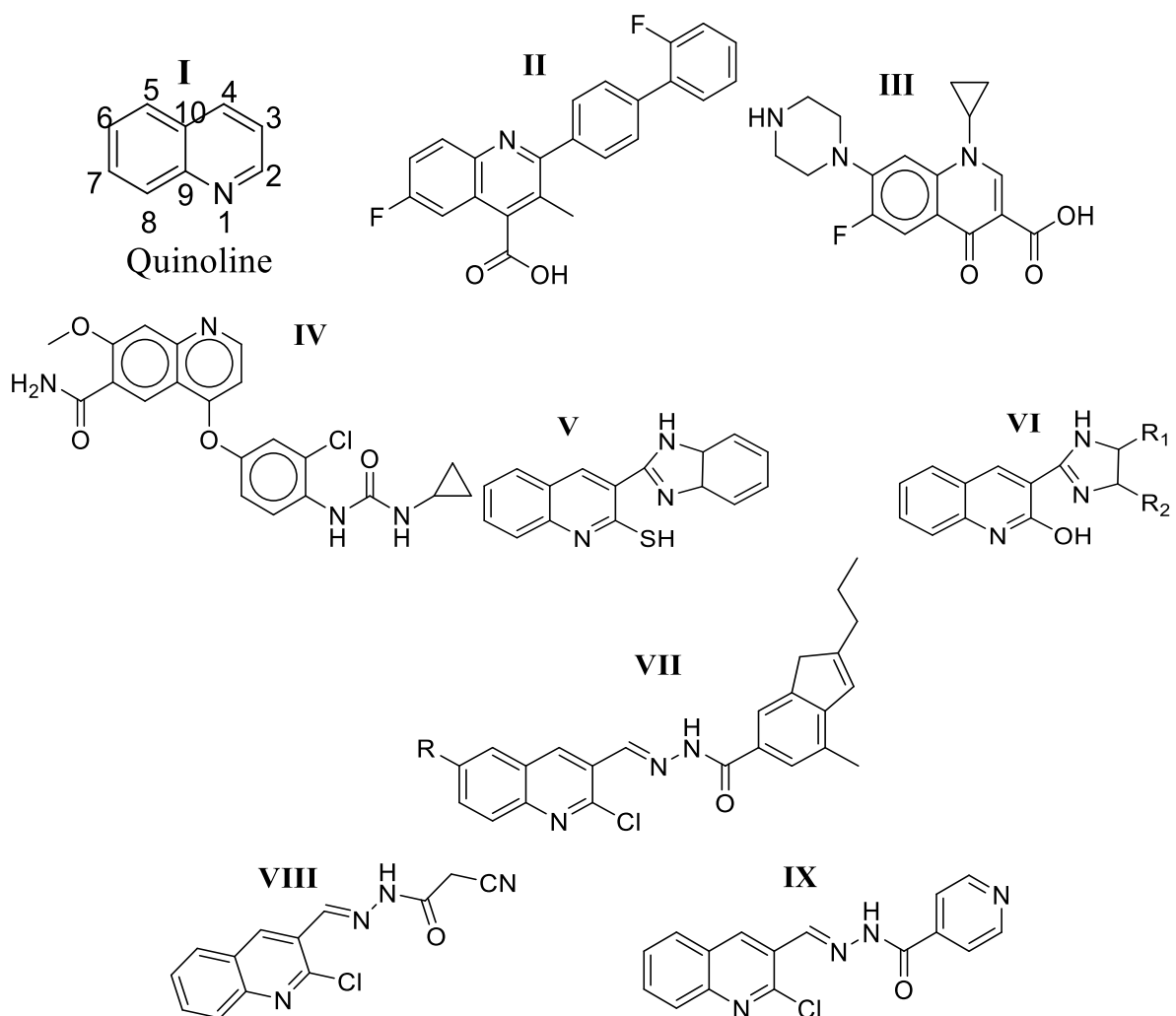


Figure 5. Quinoline motif structures for different biological activities

Schiff bases (Imines) are important class of compounds accomplished via condensation reaction of aromatic aldehydes or ketones with primary amines resulting in the introduction of imine or azomethine moiety. Schiff bases have been ubiquitously present in many biological systems including pyridoxal phosphate enzymes. Schiff bases in heterocyclic chemistry have been productive area of research for a substantial period. Schiff bases of 2-chloroquinoline-3-carbaldehyde have attracted attention mainly due to potential application as anti-microbial, anti-malarial, and anti-inflammatory agents. Benzimidazole Schiff bases have been employed as ligands for many coordination complexes. Many benzimidazole moieties have been part of numerous drugs, specifically 7-methyl-2-propyl-3*H*benzoimidazole-5-carboxylic acid forms

an important structural core of anti-hypertensive drugs (Hamama *et al.*, 2018; Mallandur *et al.*, 2017).

2.4. Silver nanoparticles (AgNPs)

Since early times, heavy metals such as copper, gold, and silver have been extensively used for the control and treatment of infectious diseases (Tomić and Vuković, 2022). The metal-based nanoparticles such as silver, gold, copper, iron, zinc, platinum, and so on, received much attention in medicine. Currently, nanoparticles can be synthesized and improved by modified the chemical groups, which help to bind the antibodies. Noble metal nanoparticles (Ag, Au, Pt) have been used for several biomedical applications such as anticancer, radiotherapy enhancement, drug delivery, thermal ablation, antibacterial, diagnostic assays, antifungal, gene delivery, and many others. The noble metal nanoparticles have some unique properties that make it more valuable. Metal nanoparticles can be functionalized with a variety of functional groups, such as peptides, antibodies, RNA, and DNA, to target different cells along with potential biocompatible polymers, for example, polyethylene glycol. Some important nanoparticles along with their biological applications to target biological cells such as cancer cell includes skin cancer, brain cancer, antimicrobial agent and antioxidant agent (Burlec *et al.*, 2023; Chandrakala *et al.*, 2022; Skłodowski *et al.*, 2023).

Out of these, Ag is the foremost metal that has been commonly used because of its well-known antimicrobial properties, and hence as an antimicrobial agent, it is always preferred in medical applications. Ag also shows potential activity against antibiotic-resistant organisms, which is one of the major issues in public health care. Nowadays, the place of metallic Ag has been taken over by Ag nanoparticles (AgNPs). Moreover, several studies performed in the past few years on AgNPs have proved their potential as a significant antimicrobial agent, which can combat various infectious diseases and be used as novel nano-medicine (Brandelli, 2015; Khaldoun *et al.*, 2024; Kotrange *et al.*, 2021).

In fact, the activity of any noble metal nanoparticle including Ag mainly depends on its colloidal stability. Therefore, the stability of metallic nanoparticles is normally achieved by

surface modification, which is relatively easy and can be accomplished through non-covalent and covalent interactions. Such modifications not only allow the replacement of potentially toxic initial stabilizing agents, but specific biological targeting and their use in bio-diagnostic and bio-sensing applications are also possible (Guerrini *et al.*, 2018; Klębowski *et al.*, 2018; Sanità *et al.*, 2020; Tim *et al.*, 2022).

There are different methods for the preparation of metal nanoparticles like chemical or photochemical methods. By using reducing agents, the metal ions are reduced to the metal nanoparticles. These have a high surface area and have the good adsorption ability of small molecules. They are widely used in different research areas, environmental and bio-imaging studies. Not only a single nanoparticle but also the mixing of two or more nanoparticles with the size control can also be achieved. By doping different metals even, the rare earth metals can change the main element characteristics. By doping different elements in different constitutions their properties also get vary (Altammar, 2023; Jamkhande *et al.*, 2019; Szczyglewska *et al.*, 2023).

The nanomaterial that play a vital role in different applications in medical science commonly occur in some basic forms. Based on the nano concept, nanomedicine is also gaining more attention by making better diagnosis and developing different ways of treatment using nanoparticles in many diagnostic devices. These metal nanoparticles exhibit resonance electron oscillation known as localized surface plasmon resonance (SPR). The ability to integrate the noble metal nanoparticles into biological system with nontoxicity has impacted significantly medicinal and biological research. Among noble metallic nanoparticles, the gold nanoparticles got much attention due to its distinctive low toxicity, ease of preparation, and favorable attachment with biological molecules (Joseph *et al.*, 2023; Yaqoob *et al.*, 2020; Zhu *et al.*, 2021).

In recent years, the concern for impact of nanoparticles on human and environment is prevailing. This kind of attention is arising in scientific community due to the release of nanoparticles from nano-based products into the environment (Kumah *et al.*, 2023a). The major concern among various nanoparticles is regarding the metallic nanoparticles in general

and AgNPs in particular, as they are being used mostly as a powerful antibacterial agent (Kumah *et al.* 2023b). The mode of their antibacterial activity and toxicity remains to be completely unclear and disputed. AgNPs are being reported to be used in both naked and capped form. Therefore, there is need of data regarding the effect of capping on their toxicity. Recent studies led by Rani *et al.* (2022) have compared the toxicity of 8 and 38 nm sized bare and PVP decorated AgNPs on the nematode *C. elegans* (Rani *et al.*, 2022). The study claimed that silver nitrate (AgNO_3) and AgNPs have similar toxicities while the coating with PVP reduced the AgNPs toxicity. However, AgNO_3 , bare AgNPs and PVP-AgNPs (8 nm) induced oxidative DNA damage, but they were not found to form the polymerase-inhibiting DNA lesions either in nucleus or in mitochondria (Bruna *et al.*, 2021; Hu *et al.*, 2018; RBenjamin *et al.* 2008). There are many reports stating the exposure of NPs to the aquatic organisms through water bodies. If ingested these NPs, it can accumulate in the digestive system thereby affecting its growth. Furthermore, it is also well known that the hardness of water also contributes to greater extent on the bioavailability and toxicity of nanoparticles. In this concern, (Oliver *et al.*, 2014) have characterized the effect of water hardness and humic acids on the bioaccumulation and toxicity of diet borne PVP-coated AgNPs on the freshwater snail *Lymnaea stagnalis*. Conclusively, they found that both the hardness of water and humic acids did not affect the bioaccumulation and toxicity of Ag from PVP-AgNPs but affected their interactions with biological membranes. The impact of citrate coated AgNPs exposure on rainbow trout (*Oncorhynchus mykiss*) gill function was also examined based on the content of the media. The authors claimed that the stronger agglomeration of citrate coated AgNPs is linked to their specific particle toxicity and damage to the gill lysosomal membrane (Corsi *et al.*, 2022; Yue *et al.*, 2016). Otherwise, while performing the study on Zebrafish (*Danio rerio*) embryos, it has been demonstrated that the toxicity of AgNPs is largely mediated by the Ag^+ ions ((Johnson *et al.*, 2022; Quevedo *et al.*, 2021). Therefore, these results imply that the coating of nanoparticles and their sizes play vital role in deciding the degree of toxicity (Brandelli, 2015).

Nanomaterials are still garnering attention at the scientific and commercial level. The properties of nanomaterials were found to be size-dependent, and this contributed to valuable chemical and physical properties. Recent developments have resulted in the improvement in

the modeling and designing of different medical and biological tools and applications.. Furthermore, some nanomaterials are engineered such as the modification occurring in case of titanium oxide, silver oxide, copper, zinc, and so on, with some other materials. Some of them are also found in different commercial production zones, such as production of sunscreens and stain-resistant clothing, and used for investigative purposes in pharmaceuticals, diagnostic kits, imaging, magnetic resonance imaging (MRI), drug delivery, and in many more medical equipment and procedures (Beyth *et al.*, 2015; Siddiqi *et al.*, 2018; Yaqoob *et al.*, 2020).

The nanomaterials applications as shown in (Figure 6) that the first category of nanomaterial comprises the pure form of metal-based nanoparticles, which are also called metal nanoparticles (e.g., silver, copper, gold, and titanium, platinum, zinc, magnesium, iron, and alginate nanoparticles). The other types of nanomaterials are the metal oxide nanoparticles (nanoparticles), that is, titanium dioxide, silver oxide, zinc oxide, and so on. The doped metal/metal oxide/metal nanomaterials are considered as another type of class among nanomaterials. Moreover, metal sulfide and metal organic frameworks (MOFs) nanomaterials have also concerned excellent interest because of their promising properties and applications in different biological fields. For example, AgS, CuS, FeS nanoparticles, Zn-based MOF, Cu-based, Mn-based MOF, and so on as shown (Figure 6) are very commonly used in various medical applications such as drug delivery and antimicrobial activities (Ahmad and Habib, 2023; Bairagi and Reza, 2023; Reza *et al.*, 2023; Yaqoob *et al.*, 2020).

The silver nanoparticles have been proven most useful because they have excellent antimicrobial properties against lethal viruses, microbes/germs, and other nucleus containing microorganisms. These nanoparticles are certainly the most extensively utilized material among all. Thus, it has been used as antimicrobial agent in different textile industries. The noble metal nanoparticles are considered as more specific and multipurpose agents with a diversity of biomedical applications considering their use in extremely sensitive investigative assays, radiotherapy enhancement, gene delivery, thermal ablation, and drug delivery. These metallic nanoparticles are also considered to be nontoxic by the scientific community in case of gene and drug delivery applications. Moreover, metallic nanoparticles can offer diagnostic and therapeutic possibilities simultaneous. Ceramics are another class of metal oxide

nanoparticles that is currently emerging as antimicrobial agents for diagnostic purpose, in drug delivery devices, and agent in different pharmaceutical and medical treatments (Hajipour *et al.*, 2012; Morales-avila *et al.*, 2017; Salleh *et al.*, 2020).

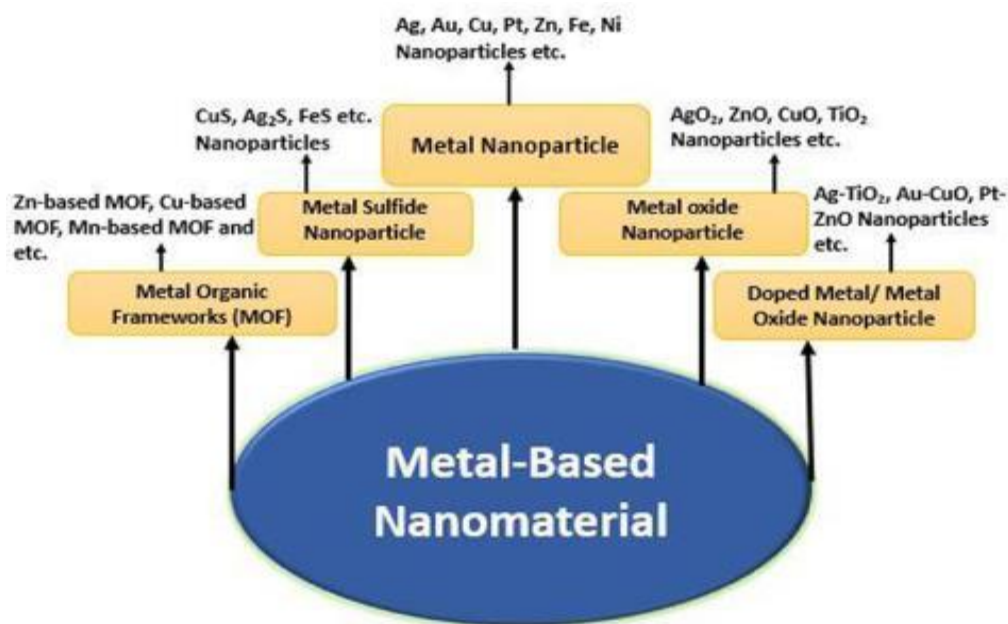


Figure 6. Different types of metal-based nanomaterials.

Owing to their high antibacterial properties, nanoparticles of silver, oxides of zinc, titanium, copper, and iron are the most used nanoparticles in antimicrobial studies. Furthermore, these nanoparticles have been used to deliver other antimicrobial drugs to the site of pathological process. Delivery of antibiotics by nanoparticles to the site of infection is a promising therapy particularly for controlled release of drugs, which in turn decreases the dose required to achieve a clinical effect. For this purpose, biodegradable nanoparticles have been used mostly. Recent studies have revealed that metallic nanoparticles, such as gold nanoparticles, may also serve as a drug-delivery system. Gold nanoparticles are of great interest as drug and gene carriers. Furthermore, since gold nanoparticles can undergo a strong plasmon resonance with light, they are used for photo-activated drug release (Kong *et al.*, 2017).

2.5. Phytochemical synthesis of Silver nanoparticles

The two most common approaches to synthesis of nanomaterials are the “top-down” and “bottom-up” methods. The top-down approach applies physical methods to change bulk materials into nano size. Although the top-down approach enables us to produce large-scale nanomaterials in a short time, a lack of control over morphology, size, and size distribution are considered major drawbacks. In contrast, the bottom-up approach enables the production of mono dispersed nanomaterials in very narrow and homogeneous size distribution with certain morphology, but nanomaterials production yield is quite low when compared to the top-down approach. In other word, the chemical and biological methods are commonly used in nanomaterial synthesis methods in the bottom-up approach (Al-Harbi and Abd-Elrahman, 2024; Paramasivam *et al.*, 2021).

Green synthesis of metal NPs has an advantage over conventional chemical and physical methods. In the recent times, green method for the synthesis of nanoparticle has become an area of utmost interest (Ying *et al.*, 2022). The conventional chemical method is expensive and requires the use of chemical compounds or organic solvents as reducing agents; those chemicals can involve various toxic chemicals be a reason for various health issues due to their toxicity. In other hand, green synthesis method is free of contaminants thus favorable for biological applications where purity is a matter of concern in different application. The use of toxic organic solvent during the synthesis and on the surface in the chemical synthesis method restricts their applications in clinical and biological fields of NPs (Ying *et al.*, 2022). Plant extract synthesis nanoparticle synthesized are more stable and biocompatible due to the process of coating with biogenic surfactants or capping agents and they also have reducing environmental impacts, exhibit variations morphology of nanoparticles and have a faster rate of synthesis in comparison with the nanoparticle produced by other materials. The faster rate of synthesis is due to the biological capacity to proceed as a catalyst for reactions in aqueous media at standard temperature and pressure (Kulkarni *et al.*, 2023; Singh *et al.*, 2023). The general mechanism of green synthesis nanoparticle in biological synthesis of NPs that the plant/microorganism extracts is simply mixed with a metal salt solution at room temperature. The synthesis process completes within hours. NPs synthesis using different plant extracts and

few reproducible microorganisms are for reducing and capping agents of metals (Miu and Dinischiotu, 2022; Singh *et al.*, 2023).

Recently, modern treatment modalities for different diseases have been developed using biotechnology. Metal nanoparticles (NPs) such as gold NPs, silver NPs (AgNPs), and zinc oxide NPs showed antimicrobial activities against several pathogens, enabling an alternative approach for controlling disease outbreaks. However, safety concerns have been raised concerned over the usage of AgNPs due to their potential side effects. Oral administration of chemically synthesized AgNPs in *Labeo rohita* leads to bioaccumulation in gills, liver, and muscles. Moreover, a marked alteration in hematological parameters and pathological damage in tested tissues were observed. Therefore, new bioactive compounds for safe and effective antimicrobial therapy should be considered. Biosynthesized AgNPs are supported to have a better level. Additionally, quinoline and its derivatives exhibit a variety of biological and pharmacological activities, including antioxidant, antidiabetic, immunomodulatory, anti-inflammatory, and neuroprotective effects (Ayala *et al.*, 2023; Rajkumar *et al.*, 2016; Shobana *et al.*, 2021).

According to Essawy *et al.*, (2021) reported that the effect of biosynthesis of AgNP on the enhancement of natural product/phytochemicals as antimicrobials such as rutin and heliomycine. These moderate antibacterial effects of both rutin and heliomycin could be due to their poor solubility in the aqueous phase. The addition of biosynthesized AgNPs to rutin and heliomycin improves their bioavailability for a couple of reasons: First, the electrostatic interaction between positive AgNPs and negative electron density on natural phenolic compounds, and second, the larger surface area of AgNPs increases the binding capacity to rutin and heliomycin molecules that potentiate their cellular uptake and hence their overall efficacy against infectious bacteria. This hypothesis is built on the existing evidence about the additive effect of AgNPs with phenolic plant extract in which an AgNPs and curcumin combination gave a better antibacterial effect than the individual treatment (Çalhan and Gundogan, 2020; Chikkanayakanahalli *et al.*, 2021; Eleonora *et al.*, 2022a).

Gold and silver-derived metallic nanoparticles (MNPs) have been shown to operate as transportation vehicles for antimicrobial peptides (AMPs) and their conjugates, in which the functionalized nanoparticles are produced by pairing or adhesion of AMPs on the MNP surfaces. These MNPs cause membrane disruption by binding to microbial surfaces through electrostatic interactions due to the large surface area and high surface charges. Considering that most nanoparticles can concentrate in the liver, spleen, and lymph nodes, use of noble metals as resistant compounds to oxidation and erosion is an excellent option for nanoparticle production and toxicity alleviation. Silver nanoparticles (AgNPs) have shown strong antibiotic properties by disrupting the microbial cell walls and membranes. It is believed that the silver-based MNPs can attach to the negatively charged microbial membranes, resulting in a greater positive zeta potential and subsequently membrane rupture and permeation. Interestingly, recent studies have shown that the synthesized AMPs-silver nanoparticles (synthesized using plant extract and loopful spore methods) are potent in killing mycobacteria, while the conjugates displayed no cytotoxicity or DNA damage on the macrophages from the infected subjects. Macrophages here actively endocytosed fluorescein isothiocyanate-labeled conjugates causing in nitric oxide independent intracellular killing of mycobacteria (Çalhan and Gundogan, 2020; Eleonora *et al.*, 2022b; Essawy *et al.*, 2021).

2.6. Decoration of metal complexes with nanoparticle

Several transition metal complexes have been developed for treatment of human diseases. Notable examples include cisplatin and its analogues to treat a variety of solid tumor cancers, gold (I) salts and auranofin that are used clinically for arthritis, and silver compounds that are effective for infectious diseases. However, the medical applications of these metal complexes usually encounter obstacles such as low solubility, low stability in physiological conditions and/or acquired/intrinsic resistance. As a result, a higher drug dosage is required for effective treatment, but this is usually accompanied by severe side effects on non-disease tissues owing to lack of specific delivery. Recent advance in nanotechnology offers an alternative approach to resolve many of the challenges in developing efficacious, biocompatible, nontoxic and nonimmunogenic biomaterials/medicines with controlled-release properties and enhanced specific accumulation in targeted cells/tissues, thereby achieving higher treatment efficiency

with lower side effects. Nanocrystalline silver dressings are used for antimicrobial protection in burn-wound treatment (Lok *et al.*, 2014; Ayukekbong *et al.*, 2017).

Recently, some advances have been made in nanomedicine studies to develop new biomedicine fields to overcome several limitations of multiple drug resistance and side effects in normal tissues, and enhance the efficacy, potency and the target site of the drugs (Marinova and Tamahkyarova, 2024). The integration of biological molecules with metallic nanoparticles creates hybrid systems that drive innovations in cell markers, biosensing, bioimaging, and targeted drug delivery. One promising approach involves coating metal nanoparticles with Cefixime was reported. Using a solution-based method, chemically modified silver nanoparticles (CX-AgNPs) were synthesized, with cefixime serving as both a stabilizing and reducing agent. The CX-AgNPs were systematically evaluated for their selectivity and sensitivity towards Hg(II) ions. Moreover, compared to their organic precursor, CX-AgNPs demonstrate enhanced potency against *S. pyogenes*, indicating their potential in targeted biomedical applications (Rasheed *et al.*, 2018; Baquero, 2021).

Drugs conjugated with nanoparticles are considered as efficient drug delivery systems that suffer from poor bioavailability for enhanced activity to treat infections, while some nanoparticles have also been used for bacterial detection Nano-metal particles have been approved as efficient chemical therapy owing to their high surface energy. Among all metal nanoparticles, silver has showed great biological impact (Masoud *et al.*, 2021). One promising route being studied currently for improving the synthesis and efficacy of silver nanoparticles is the incorporation of organic compounds and its complexes. For example, quinazolinone is a highly stable nitrogen-containing heterocyclic scaffold used to generate antibacterial drugs. Benzoxazine is a bicyclic compound containing an oxazine ring attached to a benzene ring that acts as a basis for the synthesis of other organic ligands such as quinazolinone. Both benzothiazole, a benzene ring fused with a thiazole ring, and heptane-1,7-dioic, commonly referred to as pimelic acid, are also used as starting materials for antibacterial compounds. These organic ligands can be coated with silver, enabling their conjugation with nanoparticles (Egodawaththa *et al.* 2022; By and Neill, 2016).

On the other hand, it is being increasingly recognized that nanoparticles, especially silver nanoparticles (AgNPs), have good antimicrobial properties. Thus, conjugation of bioactive compounds with nanoparticles has been proposed to enhance the activity. The nature of interaction is closely associated with stability of the system, which is an important parameter for practical utility of the conjugates. Unfortunately, AgNPs are generally unstable in solution and hence a AgNP-bioactive compounds system having long-term stability with enhanced activity is preferable (Pal *et al.*, 2016).

In contrast to bigger particles, nanoparticles can be picked up by mitochondria and cell nuclei. These nanoparticles, once taken up by these organelles, may have the potential to cause DNA mutations. However, in recent studies, combinations of nanoparticles with antibiotics, antimicrobial peptides and essential oils have been developed as promising approaches for reducing possible toxic effects of nanoparticles to the mammalian cells. Combined use of nanoparticles with antibiotics makes it possible to reduce the toxicity of both agents towards human cells due to decreased requirement for high dosages as well as synergistically enhancing their antimicrobial activities (Hajipour *et al.*, 2012; Coque *et al.*, 2023; Dhingra *et al.* 2020).

According to Allahverdiyev *et al.*, (2011) reviewed nanoparticle decorated with antibiotic resistance that combining nanoparticles with antibiotics and other antimicrobial agents, for instance the binding reaction of amoxicillin and silver nanoparticle. Hydroxy and amido groups in the molecule of amoxicillin react easily with silver nanoparticle by means of chelation (Figure 7). Antimicrobial groups together with silver nanoparticle produce a structure made up of a silver nanoparticle core surrounded by amoxicillin molecules (Allahverdiyev *et al.*, 2011).

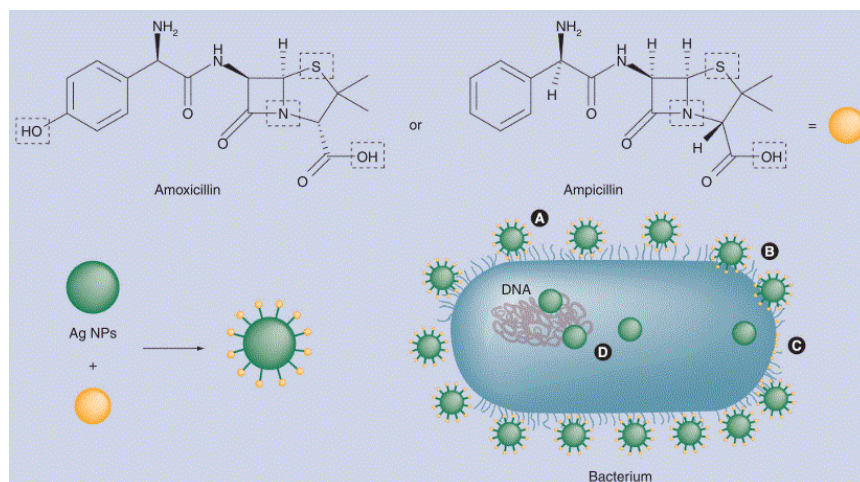


Figure 7. Mechanism of interaction between amoxicillin or ampicillin and silver nanoparticles. (A); the silver nanoparticles act as antibiotic carrier and facilitate approach of the hydrophobic antibiotic to the bacterial cell (B); penicillin antibiotics destroy the cell wall and increase its permeability for silver nanoparticles (C); silver nanoparticles prevent the DNA from unwinding (D). AgNP

Decoration/capping of AgNPs are assumed that the antimicrobial groups act on one point at the bacterial cell surface and cause more destruction by increasing the concentration of antimicrobial agents. Since silver chelation prevents DNA from unwinding, silver nanoparticles antimicrobial groups cause more dramatic DNA damage. The binding reaction between amoxicillin and silver nanoparticles attributed the enhanced activity to sulfur bridges in the molecule of amoxicillin. The most important binding sites and silver nanoparticle used as a carrier for an antibiotic. Unlike hydrophilic amoxicillin, silver nanoparticle is hydrophobic and, therefore, is more ready to interact with a bacterial membrane composed of hydrophobic phospholipids and glycoproteins. This feature of silver nanoparticle facilitates transportation of amoxicillin to the cell surface (Allahverdiyev *et al.*, 2011).

Bello-vieda *et al.*, (2018) have developed the six new copper and cobalt complexes ofazole derivatives (pyrazole, indazole, and toluene ligand) and evaluated their antibacterial activities with the combining of silver nanoparticles. The complexes have been characterized alone without the nanoparticle with FTIR, UV-visible, and TGA. FTIR of N-H, C-H, C-H, C-N, metal-Cl species functional group band intensity turned into reported given that they're found

in ligands and a metal salt. The UV-Vis spectra have been recorded to observe the electronic properties of the complexes that there are tetrahedral and octahedral complexes fashioned. Also, silver nanoparticle was synthesized with polyvinyl phenol and used Atomic Force Microscopy and Dynamic light scattering for characterization, and they report that The AgNPs agglomerated on clusters as much as 50 nm, due to the surface charge of the nanoparticles. However, the average height of the single nanoparticles was 8.35 nm, with a peak distribution between 5 to 13 nm. The evaluation of their antibacterial activities of the complexes without the nanoparticle and the combination of the complex with a silver nanoparticle that the mixture of AgNPs with the metal complexes confirmed bactericidal results at lower concentrations than within the absence of AgNPs for both Gram-positive and Gram-negative microorganism strains. The combination of metal complexes with AgNPs confirmed bactericidal activities at the same time as the free complexes and AgNPs alone simply had bacteriostatic effects. The complex mixture of silver nanoparticle activities has been increased up to 10-fold the complex alone became mentioned. The complexes combined with AgNPs showed promising activities against different bacterial strains (Bello-vieda *et al.*, 2018; Excellence and Pediatricians, 2022; Reygaert, 2018; Selgelid, 2020; Zalevskaya and Gur, 2021).

The carboxylic acid groups of the cinnamic acid Zn phthalocyanines (ZnPcs) are further used to link the phthalocyanines to the amino groups of the cysteine capped silver nanoparticles (cys-AgNPs). AgNPs are metal nanoparticles with known antimicrobial properties. AgNPs have recently been shown to improve both the triplet state properties and Photodynamic antimicrobial chemotherapy activities when linked to ZnPcs (Mafukidze *et al.*, 2019). Matlou and Nyokong, (2020a) reported on the synthesis of zinc (II) tri (tertbutyl phenoxy) mono cinnamic acid phthalocyanines complex and their linked through an amide bond to cys-AgNPs (Figure 8) and tested for their photosensitizing properties and Photocytotoxicity activity on *S. aureus*. In this case, cys-AgNPs improved the photosensitizing properties of the phthalocyanines complexes. Asymmetric Amide bond linkage of complex to cys-AgNPs and their conjugates with cinnamic as (Figure 8) recorded higher triplet and singlet oxygen quantum yield. Higher log reductions and reduction percentages for *S. aureus* were obtained

for conjugates compared to phthalocyanines complexes alone, due to their higher singlet oxygen quantum yield of the former (Matlou and Nyokong, 2020a).

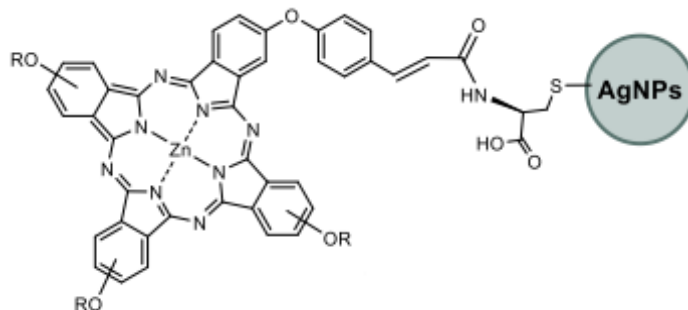


Figure 8. Structure of Zn phthalocyanines complex conjugated with cinnamic acid to cyctine AgNP.

Khoza and Nyokong, (2015) have synthesized zinc tetraamino phthalocyanine (ZnTAPc) metal complex conjugated with silver nanoparticles and immobilized on chitosan beads for the study in their photocatalytic activities. The metallic complex, metallic complex with chitosan and metal complex chitosan-AgNP had been characterized by SEM, XRD, FTIR, and TGA. The spectral shift is attributed to the changes inside the chemical environment of the ZnTAPc upon functionalization into chitosan. In addition, additionally, adjustments were discovered with the spectrum of the immobilized ZnTAPc–AgNPs result with FTIR. At 750 °C, ZnTAPc/chitosan is less stable than individual components and the presence of AgNPs improves stability consequences with TGA. The photodegradation of Rhodamine 6G becomes stronger in the presence of AgNPs whilst in comparison with the metal complexes have reported. in addition, reports have proven that coupling metallic nanoparticles which include silver or gold to a photocatalyst can also induce what is known as plasmonic photocatalysis depending on the excitation wavelength (Khoza and Nyokong, 2015).

Fratoddi and coauthors, (2019) studied the drug delivery ability of gold nanoparticles conjugated with Cu(I) complex for most cancer treatments. Gold nanoparticles (AuNPs) are the maximum versatile material in nanotechnology, with a huge variety of biological and biomedical activities, consisting of diagnostic, therapeutic, and bio-sensing applications. AuNPs had been often proposed as non-toxic carriers for drug and gene delivery applications. Copper, as an essential micronutrient in mammals, plays a pivotal role in redox-chemistry,

growth, and development, and is a key co-factor for the feature of several enzymes concerned with energy metabolism, respiration, and DNA synthesis. Novel copper-based antitumor agents had been studied in keeping with the view that endogenous metals can be much less poisonous in the direction of regular cells with admiration to cancer cells. For the reason that typically assessed mechanism of copper cellular uptake implies the reduction from copper (II) to copper (I) followed via internalization through transmembrane transporters. The gold nanocarriers are appropriate to improve the bioavailability of synthetic drugs, especially thinking about the (enhanced permeability and retention) EPR effect of AuNPs, in particular AuNPs- copper (I) complex which achieved a sluggish-release, opens the way for biological in vitro studies to explore the synergic activity of copper complexes and gold nanoparticles (Fratoddi *et al.*, 2019).

2.7. Characterization techniques for compounds.

The molecular modeling is a computer experiment that controls the system's details and basic time evolution. It can save time and cost in several scientific fields compared with experimental methods. Over the past ten years, there has been a significant increase in the computational capabilities of molecular modeling tools, which has accelerated drug discovery. It can predict the biological activity, describes the molecule's electronic properties, confirm the experimental results, and are helping to predict active compounds more efficiently. Therefore, numerous computational modeling techniques have been employed to inferior the cost and time necessary for the discovery of effective drugs. Among the most important approaches in modern drug discovery is computer-aided drug design (CADD), which consists of a broad range of theoretical and computational approaches for predicting pharmaceutical and medicinal chemistry problems. Moreover, molecular modeling, particularly docking simulation, provides information about the behavior and atomic-level interactions of molecules. It entails using computational techniques and algorithms to examine the target protein's three-dimensional structure. As a result, scientists may locate putative binding sites and refine drug candidates to improve their efficacy, specificity, and affinity (Agu *et al.*, 2023a; Ciccotti *et al.*, 2022).

The synthesized Ni(II), Cu(II), and Fe(III), metal complexes and they are characterized by FT-IR, ^1H NMR, ^{13}C NMR, and Mass spectrometry. From these instrumental analyses we find the difference between Schiff base ligands and respective metal complexes. For example, from FT-IR we differentiate the azomethine group from the peak values also in metal complexes there is an absence of phenolic $-\text{OH}$ peak value. From the ^1H NMR data, we concluded that the phenolic $-\text{OH}$ band value is absent in all type of metal complexes. From the Mass spectrometry, it was seen that the metal complexes give the $M \pm 1$ ion peak. But from the above parameters, we did not find the unit cell dimensions, measurement of sample purity, determination of crystal structures, and determination of the modal amount of minerals. Hence, for the determination of crystalline structure and other parameters like bond length, bond angle, bond type, magnetic nature, etc. we have characterized these metal complexes by other parameters such as, XRD, TGA-DTA, ESR, SEM-TEM, etc. X-ray powder diffraction is most widely used for the identification of unknown crystalline materials (e.g. minerals, inorganic compounds). Determination of unknown solids is critical to studies in geology, environmental science, material science, engineering, and biology. The XRD analysis will involve several calculations, including the characterization of crystalline materials and determination of unit cell dimensions. It will also assess sample purity, crystal structures through Rietveld refinement, and perform quantitative analysis of mineral content. Additional measurements will include dislocation density, film quality via rocking curve measurements, superlattice structures in multilayers, and film thickness, roughness, density, and grain orientation using glancing incidence X-ray reflectivity (Gopichand *et al.*, 2023; Mourdikoudis & Pallares, 2018; Prashantha & Jagadisha, 2023; Sevgi *et al.*, 2017; Zhang *et al.*, 2021).

2.8. Molecular docking

Molecular docking has now become a novel approach for the screening of synthetic bioactive compounds in drug discovery in recent years. Computer-assisted drug design is an area that is rapidly growing and has resulted in many successes. Many big pharmaceutical industries, academic resources, and research personnel are using the tools for new drug development. It is one of the most frequently used methods in structure-based drug design, due to its ability to predict the binding-conformation of compounds to the appropriate target binding site protein.

It validates the molecular mechanisms for ligands at the active site of a protein. The molecular docking approach can be used to model the interaction between a molecule and a protein, which allows us to characterize the behavior of small molecules in the binding site of target proteins as well as to elucidate fundamental biochemical processes. Molecular docking assists in the process of computer-aided drug designing by considering every possible conformation of the protein and targeted molecules. Molecular docking techniques aim to predict the best matching binding mode of a ligand to a macromolecular partner. Ligand binding to macromolecules plays a key role in medicine as a guiding mechanism in biological processes. The binding of drugs to proteins and DNA has led to vast structural studies using both experimental and theoretical methods (Agu *et al.*, 2023b; Ciccotti *et al.*, 2022).

Because DNA plays a major role in the replication and transcription of the cell, it has been a major target for antibiotics, antiviral and anticancer drugs. DNA-binding drugs are common for different diseases such as bacterial, viral, and fungal infections. The different modes of drug binding to DNA include intercalation between adjacent base pairs, and intrusion into the minor groove and the major groove. Intercalation and minor-groove binding are the predominant DNA-binding modes of small molecules. This computer-simulated ligand binding method of screening bioactive compounds is a powerful technique for investigating intermolecular interactions for rational drug design. The information gained from the docking result can be used to suggest the binding energy, free energy, and stability of complexes. The crucial advantage of molecular docking tools is that it requires less investment in resources and time in comparison to *in vivo* lab studies and important to support the *in vitro* biological studies (Alotaibi and Momen, 2019; Santos and Lamers, 2020).

CHAPTER THREE

3. MATERIAL AND METHODS

3.1. Materials

3.1.1. Chemicals and Reagents

In this study, the following chemicals and reagents were used. Silver nitrate (AgNO_3 , 99.8%, Loba Chemie, India); ferric chloride (FeCl_3 , 99% Loba Chemie, India); Muller-Hinton Agar (MHA, Merck chemical company), chloroform (CHCl_3 , 99.9% Loba Chemie, India), sulphuric acid (H_2SO_4 , 98% Loba Chemie, India), sodium hydroxide (NaOH , 98% Loba Chemie, India), ascorbic acid ($\text{C}_6\text{H}_8\text{O}_6$, 99.9% Loba Chemie, India), hydrochloric acid (HCl , 37% Loba Chemie, India), DPPH ($\text{C}_{18}\text{H}_{12}\text{N}_5\text{O}_6$, 99%, Sigma-Aldrich, Germany), L-phenylalanine ($\text{C}_9\text{H}_{11}\text{NO}_2$, 98.5%, Loba Chemie, India), and L-histidine ($\text{C}_6\text{H}_9\text{N}_3\text{O}_2$, 1.449 g/cm³ Loba Chemie, India). Ethanol absolute (99.9% Finkem), 2,2'-thiodianiline, (98%, Japan), 7-chloro-2-hydroxyquinoline-3-carbaldehyde. Acetic anhydride (99.8% Loba Chemie, India), acetic acid glacial (99.5% Loba Chemie, India), N, N-dimethyl formamide (DMF) 99% Loba Chemie, India), methanol (99.5% Loba Chemie, India), n-Hexane (99% Loba Chemie, India), dichloromethane (98% Loba Chemie, India), Ethyl acetate (99.5% Loba Chemie, India), chloroform (99% Loba Chemie, India), zinc chloride (ZnCl_2 , 98%, Loba Chemie, India), L-Ascorbic acid (99% Loba Chemie, India), dimethyl sulphoxide (DMSO, 99%, Loba Chemie, India). All the chemicals and reagents were of analytical grade and used with no further purification.

3.1.2. Plant selection and collection

In this study, selection of plants was based on ethnobotanical literature and information obtained from local healers prescribing folk medicine. During selection, priority is given to species endemic to Ethiopia, broad spectrum traditional use, and not studied for synthesis of AgNP as capping and reducing agents. For this, the leaf extract of *lippa abyssinica* was

chosen. This plant leaves collected from the Guto Gida district, East Wollega Zone, Oromia State, Ethiopia, located 335 km from Addis Ababa at Latitude: 9° 04' 60.00" N and Longitude: 36° 32' 59.99" E. This plant with voucher No. HG-2023 was identified and authenticated by the Department of Biology, Wollega University. The Voucher specimen was deposited in the herbarium of Department of Biological, Wollega University, Ethiopia.

3.2. Apparatus and Instruments

3.2.1. Physicochemical measurements

A better understanding of the behavior of metal complexes can be achieved by the study of their physical properties, which includes color, solubility, melting point, and conductivity measurements, and the chemical properties of the complexes, such as a chloride test using AgNO₃ salt solution. These are essential to gain supportive information regarding the ligand and complex formation and determine their characteristics during the chemical processes. The different chemicals have their own unique physical properties, and their detailed study helps in designing the mechanism of the reaction and helps in predicting the nature of the compounds.

3.2.2. The UV-Vis spectra

The optical properties and band gap energy of the biogenic synthesized AgNPs-His, ligand, the metal complexes and the complexes decorated with AgNPs solutions in DMSO were characterized by The UV-Vis Shanghai Drawell Scientific Instrument spectrophotometer. The spectrum was recorded in the wavelength range of 200–800 nm.

3.2.3. ¹H-NMR and ¹³C-NMR spectroscopic

The ¹H and ¹³C NMR spectra of the ligand and its complexes with Zn(II) metal ion were recorded with an Agilent 400/54 Premium spectrometer 9.4 T shielded, working at 400 MHz DMSO for ¹H and 101 MHz DMSO for ¹³C. For the analyses, the samples were dissolved in

DMSO at a concentration of 10 mg mL⁻¹. The chemical shifts (δ ppm) were calibrated to residual solvent signals.

3.2.4. Fourier transform infrared spectroscopy (FTIR)

The FTIR spectra were recorded on PerkinElmer Spectrum 65 FT-IR Spectrometer. The FTIR spectra were collected at standard spatial resolution in the range of 400-4000 cm⁻¹. The samples were made using the KBr pellets technique (Suresh et al., 2016).

3.2.5. Scanning Electron Microscope (SEM) with energy dispersive X-ray (EDX)

SEM-EDX analysis was carried out using Scanning Electron Microscope EVO18 (CARL ZEISS) and Energy Dispersive X-ray-Spectrometer Quanta 200 with X Flash 6130. The oven-dried biogenic AgNP-His and its decorated with the ligand and Cu(II) complexes were loaded onto the stub with carbon tape fixed and coated by sputtering with gold.

3.2.6. Transmission electron microscope (TEM)

Transmission electron microscopy was recorded at 200 kV on TEM, HF5000, Hitachi High-Technologies, Japan equipped with an EDS energy dispersive system was used. HRTEM technology, Resolution: 0.14 nm lattice, 0.12 nm point-to-point, Magnification Range: 4,000x – 1,5000,000x, High dispersion diffraction camera length range: 4 to 80 m. Specimen Tilt range: +/-20 degrees (for UHR configuration), +/-30 degrees (for HR and HC configuration). TEM images of biogenic AgNPs were processed with the ImageJ software to obtain the diameter size distribution of the particles.

3.2.7. Thermogravimetry

Using the Thermal Analysis Instrument of the SDT-Q600 analyzer (TA Instruments), the thermal stability of the Cu(II) complex was examined. This device produced differential thermal analysis (DTA) and synchronized thermogravimetry analysis (TGA) curves. An

alumina sample container was filled with a sample mass of roughly 7.0 mg, and a heating rate of 10 °C min⁻¹ was used at an airflow of 100 mL min⁻¹.

3.2.8. Powder X-ray Diffractometer (pXRD) Analysis

An X-ray diffractometer (Shimadzu Corporation (Japan); XRD-7000 X-ray diffractometer) was used for the XRD analysis to study the crystallographic nature of the biogenic AgNPs. The Cu-K α radiation source was used at a wavelength of 1.5406 Å and a scanning angle 2 θ in the scanning range of 10° to 90° on an instrument operating at a voltage of 45 kV and a current of 40 mA.

3.2.9. Time dependent (TD)-DFT analysis of the ligand and Zn(II) and Cu(II) complexes

Density Functional Theory (DFT) calculations employing B3LYP (Becke 1993; Lee *et al.*, 1988; Stephens and Devlin, 1994) hybrid functional together with 6-311++G(d, p) basis set (Krishnan *et al.*, 1980) for the light atoms and the Los Alamos National Laboratory 2-double-zeta (LanL2DZ) pseudo potential for the copper and zinc atom (Hay and Wadt, 1985) were employed using the Gaussian 16 program package (Frisch *et al.*, 2016). The polarizable continuum solvation model in its integral equation formalism together with DMSO as a solvent was used to account for solvent effects. Dispersion correction (Grimme, 2002) was employed to account for non-bonding interactions. Vibrational frequency calculations were performed to ensure that the optimized geometries are real minima without imaginary frequencies (Appendix 20). The time-dependent DFT (TD-DFT) was used to calculate absorption energies at the same level of calculations.

Quantum chemical descriptors were calculated at the same level of theory. The HOMO and LUMO energies were used to predict the quantum mechanical descriptors: band gap ($E_g = E_{\text{LUMO}} - E_{\text{HOMO}}$), electronegativity ($\chi = -\frac{1}{2} (E_{\text{HOMO}} + E_{\text{LUMO}})$), electronic chemical potential ($\mu = \frac{1}{2} (E_{\text{HOMO}} + E_{\text{LUMO}}) = -\chi$), global chemical hardness ($\eta = \frac{1}{2} (E_{\text{LUMO}} - E_{\text{HOMO}})$), global softness ($\sigma = 1/2\eta$), global electrophilicity index ($\omega = \mu^2/2\eta$), and nucleophilicity index ($Nu = 1/\omega$) of the compounds (Bitew *et al.* 2021; Ismael, Rabia, and Abdou 2020).

3.3. Experimental procedures

3.3.1. Plant material preparation and extraction

The preparation of *Lippia abyssinica* leaf extract was conducted using reported procedures with slight modifications (Arya *et al.*, 2018). The fresh leaves of *Lippia abyssinica* were collected and washed with tap water several times, followed by rinsing with double distilled water to remove any dust particles. The cleaned leaves were dried in open air under shade. The dried leaves were ground into a fine powder using an electric blender and kept for extraction at ambient temperature. The extraction was carried out by taking 20 g of the powdered plant leaves in a 500 mL conical flask containing 400 mL of distilled water. It was covered with aluminum foil to prevent the effect of light on the solution. The mixture was stirred for 30 min and then heated at 60 °C with continuous stirring on a hot plate for 1 h. After cooling down to room temperature, the solution was filtered with Whatman No.1 filter paper to get a clear extract solution as shown in Figure 9. The filtrate was stored at 4 °C for further experiments.

3.3.1.1. Phytochemical screening

A preliminary phytochemical screening of the aqueous leaf extracts was performed using reported techniques for phenolic, alkaloids, Flavonoids, Terpenoids, Steroids, tannin and Saponins (Gul *et al.*, 2017; Ram and Sinha, 2015; Senguttuvan *et al.*, 2014).

Phenolic test: The Ferric chloride test method, which involves adding 2 mL of a 5 % solution of FeCl₃ to 1 mL of *Lippia abyssinica* leaf extract in a test tube and shaking the mixture together before standing the test tube upright in a rack of test tubes, was the technique used to determine phenol qualitatively. The reaction mixture had a black color, indicating that phenol was indeed present.

Alkaloids Test (Mayer's test): The mixture of 3 mL of *Lippia abyssinica* leaf extract and 3 mL of 1% HCl solution was heated for 20 min in a water bath. Mayer's test was run using the reaction mixture that resulted after being cooled to room temperature. Drop by drop, 1 mL of Mayer's reagent (1.36 % HgCl₂ and 5% KI) was added to the reaction mixture after cooling.

The existence of alkaloids was evidenced by the precipitation of a greenish-colored cream precipitate.

Flavonoids Test (Alkaline reagent test): 3 mL of extract was added to 1 mL of a 10 % NaOH solution in the test tube. A bright yellow hue was developed, which supports the presence of flavonoids. By dropping diluted HCl the color gradually vanished.

Terpenoids and Steroids Salkowski test: The test tube was filled with 5 mL of extract, 2 mL of chloroform, and 3 mL of concentrated sulfuric acid, and it was thoroughly shaken to create a layer. The interface took on a reddish-brown color, indicating the presence of terpenoids. The lower surface is red, indicating the presence of steroids.

Tannins test:

- A. Ferric chloride test:** When 1 mL of distilled water and 2 drops of concentrated ferric chloride solution were added to 0.5 mL of extract in a test tube, blue-black coloration was noticed, indicating the presence of tannins.
- B. Lead acetate test:** When 0.5 mL of extract in the test tube solution was added to a 10% lead acetate solution, white precipitation was observed, confirming the presence of tannins.

Saponins-Frothing test: In the test tube, 0.2 mL of the extract was shaken with 5 mL of distilled water. Saponin is detected through foaming.

3.3.2. Biogenic synthesis of silver nanoparticles

3.3.2.1 Optimization of various parameters for silver nanoparticle synthesis

The parameters (time, pH, temperature, concentration of silver nitrate, and plant extract) were optimized for the rapid and maximum synthesis of AgNPs. The pH was maintained at 2.99, 5.58, 6.99, 8.34, 10.34, and 12.01 and optimized with 0.1 N hydrochloride acid (HCl) and 0.1 N sodium hydroxide (NaOH). The reaction was monitored at 0 min, 30 min, 1 h, 2 h and 4 h for optimal production of AgNPs. The synthesis of silver nanoparticles was monitored at

various incubation temperatures (10, 20, 50, 80, and 100 °C). The 50 mL aqueous solution of AgNO₃ was used for optimized at different concentrations (2, 4, 8, 12, and 16 mM). Similarly, the concentration ratio of plant extract and AgNO₃ was optimized by increasing the concentration of plant extract (2, 4, 6, 8, and 10 mL) in a 2 mM AgNO₃ solution. The synthesis was observed at various plant extracts to silver nitrate solution ratios. The absorbance of nanoparticles was noted within the range of 200–800 nm by a UV–visible spectrophotometer.

The optimal volume (10 mL) of extract (1/20 weight/volume) was combined with the optimal concentration (2 mM) of AgNO₃ solution for the reduction of Ag ions at an optimized temperature and reaction time (80 °C and 2 h). The reaction was carried out in a dark by aluminum foil covering with. The solution was continuously homogenized while being stirred at 10,000 rpm at a desired temperature. A double-beam UV-Vis spectrophotometer was used to measure the reaction progress to optimize the AgNO₃ solution concentrations, reaction time, volume ratio of AgNO₃ to plant leave extract, reaction mixture temperatures, and pH effects. Following the optimization of the reaction conditions, a color change from colorless to brown was noticed, which indicated the formation of silver nanoparticles. The product was separated from the mixture using high-speed centrifugation for 20 minutes at 10,000 rpm. Then, the synthesized AgNPs powders were rinsed with 20 mL of distilled water and dried at 105 °C for further characterizations (Arya *et al.*, 2018).

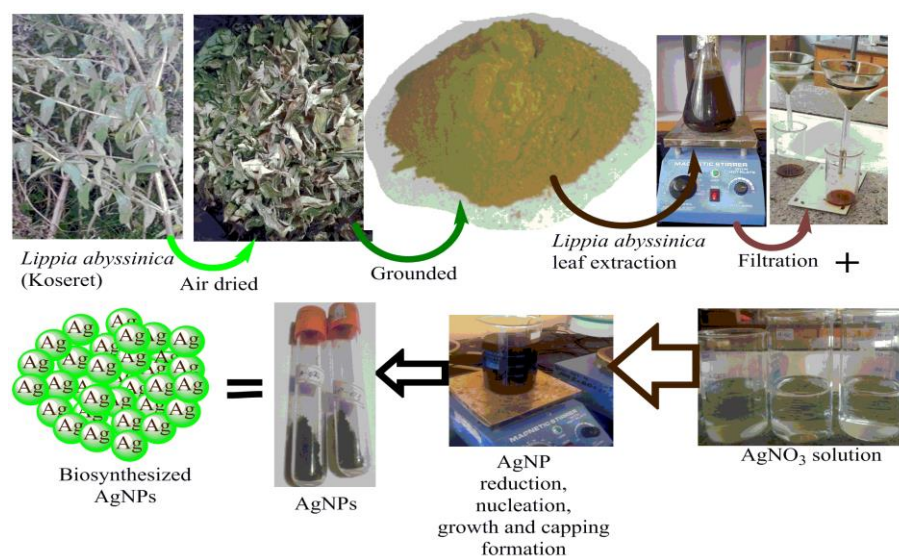


Figure 9. Schematic representation for the leaf extract-mediated biogenic of AgNPs

3.3.3. Biogenic synthesis of silver nanoparticles functionalized with histidine and phenylalanine

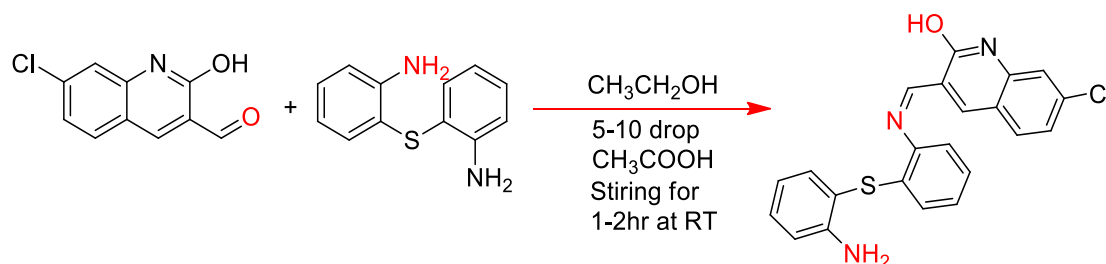
The synthesis of functionalized silver nanoparticles (f-AgNPs) was conducted using reported procedure with slight modification (Chandra and Singh, 2018). Briefly, 10 mL aqueous extract was added to 2 mM of 50 mL of aqueous solution of AgNO₃ in 100 mL volumetric flask. Subsequently, (2, 4, 8, 10 mL) for volume optimization of aqueous solution of 0.01M of histidine or phenylalanine as a surface-modifying agent was added to the reaction mixture drop-by-drop while stirring on hot plate as shown in Figure 9. The pH of the solution was adjusted to 12.01 using 1 M NaOH solution. The reaction was left to proceed for 2 h at 80 °C under constant stirring. Finally, the f-AgNPs were separated from the mixture by high-speed centrifugation at 10,000 rpm for 20 min. After centrifugation, the synthesized f-AgNPs powders were rinsed with 20 mL distilled water and dried in oven at 105 °C for further characterizations.

3.3.4. Synthesis of the Imine-Quinoline Derivative Ligand and its Zn(II) and Cu(II) complexes

3.3.4.1. Synthesis of ligand (L) (Z)-3-(((2-((2-aminophenyl)thio)phenyl)imino)methyl)-7-chloronaphthalen-2-ol.

The imine-quinoline derivative ligand, (Z)-3-(((2-((2-aminophenyl)thio)phenyl)imino)methyl)-7-chloronaphthalen-2-ol was prepared as described in Scheme 1 (mechanism is illustrated in Appendix 3) by the reaction of 7-chloro-2-hydroxyquinoline-3-carbaldehyde and 2,2'-thiodianiline in 1:1 molar ratio using previously reported procedures (Damena *et al.*, 2022a; Dinku *et al.*, 2024; Hussain *et al.*, 2014; Naganagowda *et al.*, 2014; Tapabashi *et al.*, 2021). Specifically, to a solution of 7-chloro-2-hydroxyquinoline-3-carbaldehyde (10 mmol, 1.5 g) in 15 mL of ethanol, a solution of 2,2'-thiodianiline (10 mmol, 2.2 g) in 15 mL of ethanol was gradually added while stirring. Then, the solution was stirred for 2 h at room temperature. Then 5–10 drops of glacial acetic acid were added to the mixture with constant

stirring. The reaction progress was followed by using TLC. A yellow precipitate was formed and was then filtered and repeatedly washed with ethanol. After drying, the actual yield of the product was calculated. (Melting point, 145-150 °C, yield (89.2%)) of the ligand



Scheme 1. Synthesis of imine quinoline ligand, (Z)-3-(((2-((2-aminophenyl)thio)phenyl)imino)methyl)-7-chloronaphthalen-2-ol.

3.3.4 2. Synthesis of Zn(II) complexes of ZnL and ZnL₂

The synthesis of the Zn(II) complex was carried out using the following procedure. (Z)-3-(((2-((2-aminophenyl)thio)phenyl)imino)methyl)-7-chloronaphthalen-2-ol (L) (0.220 g, 1 mmol) was dissolved in 20 mL of ethanol in round bottom flask with constant stirring on the hot plate. Then a sample of solid zinc chloride (ZnCl_2) (0.136 g, 1 mmol) was added to the hot solution for the 1:1 of the Metal to Ligand molar ratio. Similarly (0.440 g, 2 mmol) of the ligand and (0.136 g, 1 mmol) of ZnCl_2 for 1:2 of the metal to ligand molar ratio. The resultant mixture was refluxed for 3-4 h at a temperature of 60-70 °C. The reaction progress was checked using TLC to confirm the reaction completion. In both case the pale-yellow precipitates were obtained upon cooling and the products were kept at room temperature overnight. The results were filtered out, washed with cold ethanol, and dried (Damena *et al.*, 2022a; Dinku *et al.*, 2024; Hussain *et al.*, 2014; Naganagowda *et al.*, 2014; Tapabashi *et al.*, 2021). The products melting points were recorded > 200 °C and the yield of ZnL and ZnL₂ were 76.12 and 86.0% respectively, were obtained.

3.3.4.3. Synthesis of Cu(II) complexes with the imine quinoline ligand; CuL and CuL₂

Copper (II) chloride dihydrate ($\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$) (0.170 g; 1 mmol) was added to a ethanolic solution of (Z)-3-(((2-((2-aminophenyl)thio)phenyl)imino)methyl)-7-chloronaphthalen-2-ol (L) (0.220 g; 1 mmol) in 1:1 molar ratio, and ($\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$) (0.170 g; 1 mmol) and L (0.440.

g; 2 mmol) in 1:2 molar ratio of metal to ligand. The solution mixture was stirred and refluxed at 60-70 °C for 3-4 h. Then blue and brick red solutions of CuL₂ and CuL, respectively, were obtained. The precipitates were formed upon cooling to room temperature overnight. It was filtered out, washed with cold ethanol, and dried. Yield = 69.23% (for CuL) melting point > 200 °C and 75.4% (for CuL₂), melting point > 200 °C respectively (Damena *et al.*, 2022a; Dinku *et al.*, 2024; Hussain *et al.*, 2014; Naganagowda *et al.*, 2014; Tapabashi *et al.*, 2021)..

3.3.5. Synthesis of the ligand and the metal complexes decorated with silver nanoparticle

3.3.5.1. Synthesis of AgNP decorated with imine-quinoline ligand

The histidine-functionalized AgNPs (AgNP-his) was selected for decoration with the ligand and metal complexes. The selection of AgNPs functionalized with histidine over those synthesized using plant extract alone was driven by the superior properties exhibited by the histidine-functionalized nanoparticles. These AgNPs demonstrated a smaller size and enhanced antibacterial and antioxidant activities compared to AgNPs synthesized solely with plant extract. Additionally, when compared to AgNPs functionalized with phenylalanine, which resulted in larger particle sizes, the histidine-functionalized AgNPs offered more favorable characteristics for further applications (Chakraborty *et al.*, 2018; Chandra and Singh, 2018). This combination of small size and high bioactivity made AgNP-histidine an ideal candidate for compositing with imine quinoline ligands and Cu(II) and Zn(II) complexes for enhanced biological studies.

Decorated of the ligand with AgNPs was carried out by using standard reported procedure (Cai *et al.*, 2021; Mostafa *et al.*, 2023; Wang *et al.*, 2021) with slight modification. The imine quinoline ligand loaded with AgNPs using 3:7 weight ratio of AgNPs to imine quinoline ligand. 0.047 g of the metal complexes were prepared separately in 25 mL of ethanol and 0.047 g of imine quinoline was dissolved in 25 mL of ethanol then it was added drop wise to the previously prepared AgNPs with constant stirring for 6 h at room temperature. The

composite was precipitated in a mixture of acetone and ethanol, filtered, and finally dried in air.

3.3.5.2. Synthesis of decorated AgNP with Cu(II) and Zn(II) complexes

The synthesis of decorated AgNPs with metal complexes were carried out following a procedure reported elsewhere (Cai *et al.*, 2021; Mostafa *et al.*, 2023; Wang *et al.*, 2021) with slight modification time of reaction. The Cu(II) and Zn(II) complexes were loaded with AgNPs using 3:7 weight ratio of AgNPs to each metal complexes. 0.02 g of AgNPs were dispersed individually in ethanol by sonication for 40 min for each complexes. 0.047 g of the metal complexes (CuL, CuL₂, ZnL or ZnL₂) were prepared separately in 25 mL of ethanol then it was added drop wise to the previously prepared the AgNPs solution with constant stirring for 6 h. The composite was precipitated in a mixture of acetone and ethanol, filtered, and finally dried in air.

3.4. Biological activities

3.4.1. Antibacterial Activity

The antibacterial activities of imine-quinoline ligand, its pristine metal complexes and AgNP decorated counterparts were studied on Mueller Hinton agar media using an agar-well diffusion method following standard protocol ref. Specifically, 1 mL of actively developing clinical strains of gram-negative (*Escherichia coli* ATCC 25922 (E. coli) and *Pseudomonas aeruginosa* (*P. aeruginosa*)) and gram positive (*Staphylococcus aureus* ATCC 25926 (S. aureus)) (containing about 1×10^7 CFU mL⁻¹, 0.5 McFarland Standard) bacteria were dispersed on a homogenous plate using cotton swabs. Wells were created in each agar culture medium, using a sterile cork borer (6 mm in diameter) and filled in triplicate of the samples. Dimethyl sulfoxide (DMSO) and gentamicin were used as negative and positive control respectively. The culture media were kept in the refrigerator for 30 min. before being incubated for 24 h at 37 °C. Using a Vernier caliper, the diameter of the growth-free zones was measured in mm to determine the zones of inhibition (Hudzicki, 2009).

3.4.2. Minimum Inhibitory Concentration

A microplate dilution method with slight modification (Humphries *et al.*, 2019) was used to determine the minimum growth inhibition concentration (MIC) for the ligand and its Cu(II) and Zn(II) complexes. Their separate solutions were prepared in 10% DMSO in two-fold dilutions. Bacterial suspensions were prepared from Mueller-Hinton broth and 100 µL of the suspensions were added to 1.5 µL microcentrifuge tubes with 300 µL of the compounds. Microcentrifuge tubes containing sample solutions without bacterial suspension served as negative control whereas those containing bacterial suspension with various gentamicin concentrations were positive controls. The studies included tubes containing sample solution without bacterial suspension (negative control) and tubes containing bacterial suspension with various gentamicin concentrations (positive control). The tubes were incubated for 24 h at 35±2 °C. Using a UV-VIS spectrophotometer (Drawell, DU-8800R), absorbance at 630 nm was used to measure the antimicrobial activity. As a blank, the negative control was employed (Mishra *et al.* 2017).

3.4.3. Antioxidant Activity

The antioxidant activities of the imine quinoline ligand (L), and its Cu(II) and Zn(II) complexes and their decorated AgNP were assessed using a conventional protocol. Separate test tubes with standard ascorbic acid and different concentrations of the ligand (L) and its Cu(II) and Zn(II), complexes (50, 100, 150, 200, and 250 µg/mL for each) were prepared in separate test tubes from a 50 mg/mL stock solution of each the synthesized compounds. After adding 3 mL of a 0.1 mM methanol solution of DPPH to each prepared solution, the test tubes were kept in the dark and allowed to incubate for 20 min. After incubation, the absorbance of the solutions at 517 nm was measured using a UV-Vis against methanol as a blank. The standard was prepared from ascorbic acid (0.2–1.0 mg/mL) in methanol. The experiments were executed in triplication to facilitate statistical analysis. The antioxidant activity was calculated as a percentage of activity by the following equation (Eqn.1): (Molyneux, 2004).

$$\text{The scavenging Percent activity (\%)} = \frac{Ac - As}{Ac} \times 100 \dots \dots \dots \text{Equ (1)}$$

Where Ac = the absorbance of standard/control,

As = the absorbance of the sample solutions.

3.4.4. *In Vitro* Cytotoxicity assay of MCF-7 cell line

Using the 3-[4,5-dimethylthiazole-2-yl]-2,5-diphenyltetrazolium bromide (MTT) test (Chen *et al.*, 1990; Kumar *et al.*, 2018), the cytotoxicity of the substances ligand, CuL, and CuL₂ was assessed in accordance with OEM guidelines (Roche) with the concentration of (0, 6.125, 12.5, 25, 50, 100, 200 µg/mL) for each. A transparent, flat-bottomed 96-well plate was used to seed MCF-7 breast cancer cells at a density of 5×10^3 /cells well in 100 µL of growth media. For a whole day, the cells were left to adhere. In phosphate-buffered saline, or PBS (pH about 7.4), the as-synthesized compounds were dissolved. Growth medium was utilized to create doses that ranged from 25 to 0.78 µM by repeated dilution. After discarding the spent growth medium, the cells were treated for 24 hours at 37.5° C with the compounds that had been diluted with the growth medium. Using the same protocol, control cells were treated with a PBS mixed solvent at doses equivalent to the drugs included, as a positive control was cisplatin. Following the 24-h incubation period, each well received 10 µL of MTT labeling reagent (final concentration: 0.5 mg/mL), which was then added. The wells were then incubated for 4 hours at 37.5 °C in a humidified 5% CO₂ environment. The formazan crystals were dissolved by adding DMSO to each well. The optical density (OD) of the plate well was measured using a Multiskan™ FC Microplate photometer at 570 nm with microplate reader and data were maintained. Triplicated wells were conducted for each drug concentration tested. The IC₅₀ values of the compounds were calculated by the nonlinear regression analysis.

3.4.5 Molecular Docking

The binding modalities of ligand interaction at the protein's active site are defined by molecular docking experiments. Using the Molecular Operating Environment, which can be downloaded from the Protein Data Bank (<http://www.rcsb.org/pdb>) and used for the molecular docking investigation against *Staphylococcus aureus* (PDB: 2w9h) and *Escherichia coli* (PDB: 6F86), and *Estrogen Receptors Alpha* (ERα) (PDB:5GS4) a model of the ligand-protein interaction was constructed. Before docking, the protein was repaired by a series of processes. Among these steps were the elimination of co-ligands and solvent molecules, the addition of hydrogen, chain repair, and active site selection. The chemicals that were tested also went

through an energy reduction, energy adaptation, atomic charge calculation, binding energy, and inhibition constant calculation docking optimization process. In this field of study, different kinds of structures can be analyzed to ascertain the stability of H-bonds, π -alkyl, π -Sigma, and van der Waals adducts. The formula used to rate the ligand's and metal complexes' affinity for the target protein took into consideration the hydrogen bonds and binding free energy that exist between the ligand and the amino acid. To conclude, using Discovery Studio software, the conformers with the lowest binding free energies were employed to show how the compounds interacted with the active amino acids. To compare the docking results with the equivalent experimental data, gentamicin was chosen as a positive control (Meng *et al.*, 2011).

3.5. Statistical Analysis

The biological activities evaluation data with triplicated measurement were summarized as means $\pm$ standard deviation in which Graph pad prism version 5.00 was used for the analysis (Graphpad software, California, United State) (Kerr *et al.*, 2020). Groups were analyzed for significant difference using analysis of variance (ANOVA) test whether there is a significant difference among the treatment means at $P < 5\%$ ($P < 5\%$) using the Statistical Analysis Software (SAS 14.1).

CHAPTER FOUR

4. RESULTS AND DISCUSSION

4.1. General overview

In this research work, the inquiry into the synthesis of new metal complexes of Cu(II) and Zn(II) with an imine quinoline derivative ligands are presented. 7-chloro-2-hydroxyquinoline-3-carbaldehyde and 2, 2'-thiodianiline combine to create the imine quinoline derivative, which provides a NO donor atom for chelation of the metal ions. The biogenic of silver nanoparticles was carried using *lippa abyssinica* leave extract (LALE) with selected amino acids (histidine and phenylalanine) as reducing and capping agents. Furthermore, the AgNP-His was decorated with the L and its complexes of Zn(II) and Cu(II). AgNPs, L and its complexes with Cu(II), Zn(II), and the decorated AgNP-His with the ligand, **CuL**, **CuL₂**, **ZnL**, and **ZnL₂** are the synthesized compounds and their biological activities were evaluated and reported. The FTIR, NMR, UV-Vis, EDX, and microscopy studies (TEM, HRTEM, SEAD and SEM) were used to describe the characteristics of the synthesized materials.

4.2. Physical measurement

The study of physical attributes such as color, solubility, melting point, chloride test, and conductivity test might help for better understanding of the behavior of the synthesized nanoparticles, ligands, and metal complexes. The understanding of the distinct physical and chemical characteristics aid in both anticipating the composition of the compounds, and the reaction process. One of the most important physical characteristics for the synthesis of nanoparticles is the presence of changes in color.

The ligand electron cloud interacts with the transitional metal ion degenerate d-orbitals to divide them into distinct energy levels that are determined by the complexes shapes. When electromagnetic radiation is exposed to such complexes, a distinctive energy absorption and electronic transition from lower to higher energy level was recorded. The complex releases

energy in response to absorbed radiation and changes the chemical color if it absorbs electromagnetic radiation in the visible portion of the spectrum. The complexes varying range of electronic absorption, by either free electrons or by π bonding or nonbonding electrons, causes a change in color. When auxochromes are present, chromophores often exhibit electronic absorption in the UV-visible range, giving substances their distinctive color. The free electrons in metal complexes show electronic absorption, giving the metal complexes their distinctive colors.

4.2.1. Solubility test of the ligand and its Cu(II) and Zn(II) complexes

The solubility of the synthesized imine quinoline ligand (L) and its Cu(II) and Zn(II) complexes was tested with the following solvents: water, dichloromethane, methanol, ethanol, acetonitrile, dimethyl formamide (DMF), dimethylsulfoxide (DMSO), and chloroform as shown in Table 1. The results revealed that the imine quinoline ligand L and its Cu(II) and Zn(II) complexes were soluble in the aforementioned organic solvents and sparingly soluble in water.

Table 1. The solubility test of the ligand and its Cu(II) and Zn(II) complexes

Solvents	Synthesized compounds				
	L	CuL	CuL ₂	ZnL	ZnL ₂
Water	Sparingly soluble	Sparingly soluble	Sparingly soluble	Sparingly soluble	Sparingly soluble
Dichloromethane	Soluble	Soluble	Soluble	Soluble	Soluble
Methanol	Soluble	Soluble	Soluble	Soluble	Soluble
Ethanol	Soluble	Soluble	Soluble	Soluble	Soluble
acetonitrile	Soluble	Soluble	Soluble	Soluble	Soluble
DMF	Soluble	Soluble	Soluble	Soluble	Soluble
DMSO	Soluble	Soluble	Soluble	Soluble	Soluble
Chloroform	Soluble	Soluble	Soluble	Soluble	Soluble

Here, the solubility behaviour of L and its Cu(II) and Zn(II) complexes is readily soluble in organic solvents like methanol and ethanol but partially soluble in water. Several factors, such as their molecular structure, polarity, and interactions with the solvent, might influence them. Imine-quinoline ligands typically contain both hydrophobic (non-polar) and hydrophilic (polar) regions. Methanol and ethanol are polar protic solvents; water is a polar solvent. The mixed polarity and the bulky, hydrophobic quinoline structure make these ligands and complexes readily soluble in organic solvents but less soluble in water as confirmed by *in silico* analysis (Appendix 21).

4.2.2. Physicochemical characteristics of L and its Cu(II) and Zn(II) complexes

The physicochemical characteristics of the imine-quinoline ligand (L) and its Cu(II) and Zn(II) complexes reveal distinct properties as indicated in Table 2. The free ligand (L) is yellow, a typical color for quinoline-based compounds, indicating absorption in the violet-blue region of the spectrum. Upon coordination, the Zn(II) complexes, ZnL and ZnL₂ remain light yellow, reflecting the d¹⁰ electronic configuration of Zn(II), which results in no d-d transitions and light-colored complexes. The Cu(II) complexes, CuL (brick red) and CuL₂ (blue), exhibit more pronounced colors due to Cu(II) d⁹ configuration, with d-d transition and the blue color of CuL₂ being characteristic of octahedral.

The metal complexes were dissolved in DMSO for molar conductance analysis to determine their electrolytic and non-electrolytic nature. The molar conductance of the complexes were found to be 4.45, 11.05, 3.35 and 5.71 Scm²mol⁻¹ for CuL, CuL₂, ZnL and ZnL₂ respectively (Table 2), indicating non-electrolyte nature of the complexes (Chandra & Kumar, 2005). This is mainly attributed to the metal cations receive electrons from the ligand to bring a net charge balance to zero. The molar conductance results can also provide information about the proposed structures of the complexes. Also the metal complexes were non electrolyte due to low solubility and absence of anion beyond the coordination sphere (Jayaseelan *et al.*, 2016). These will decrease its ion mobility and molar conductance. The chloride test of the metal complexes also carried out by using a mole of AgNO₃ solution that there was no white precipitate in the solutions, which validate the absence of chloride ions in the solution of the

complexes, indicates chloride ions are coordinated to the metal center. All the metal complexes exhibit high thermal stability, with melting points exceeding 200 °C, significantly higher than the melting point of the free ligand (145-150 °C). This suggests stronger intermolecular forces due to metal-ligand coordination. The pH of the free ligand (L) is 6.74, indicating slight acidity, while the pH of the complexes is slightly lower, ranging from 6.23 to 6.48, suggesting that complexation with Zn(II) and Cu(II) slightly increases the acidity of the ligand's environment. This could be due to the interaction of water molecules and chloride ions with the metal center.

Table 2. The molar conductivities color and chloride test of the Zn(II) and Cu(II) complexes

Complex	Color	Molar conductance $\text{Scm}^2\text{mol}^{-1}$	Yield (%)	Chloride test	Melting point °C	pH
L	Yellow	-	89.20	-	145-150	6.74
[ZnL(H₂O)Cl₂], MF: C ₂₂ H ₂₀ Cl ₃ N ₃ O ₃ SZn	Light yellow	3.35	76.12	No white precipitate	>200	6.46
[ZnL₂Cl₂], MF: C ₄₄ H ₃₂ Cl ₄ N ₆ O ₂ S ₂ Zn	Light yellow	5.71	86.00	No white precipitate	>200	6.48
[CuL(H₂O)₂Cl₂] C ₂₂ H ₂₀ C ₁₃ CuN ₃ O ₃ S	Brick red	4.45	69.23	No white precipitate	>200	6.23
[CuL₂Cl₂] C ₄₄ H ₃₂ C ₁₄ CuN ₆ O ₂ S ₂	Blue	11.05	75.4	No white precipitate	>200	6.34

4.3. Phytochemical analysis of plant leaves

The phytochemical test of LALE was conducted using standard protocols (Gul *et al.*, 2017; Ram and Sinha, 2015; Senguttuvan *et al.*, 2014) and the result revealed the presence of secondary metabolites as listed in Table 3. These phytochemicals are capable of reducing and capping AgNPs by donating and stabilizing the formation AgNPs. The color changes against the reagents indicate the formation of different complexes due to oxidation and reduction reactions. For instance, the yellow color of the alkaloid indicates that the nitrogen or oxygen

atoms of the amide groups of the alkaloids are involved in a reaction. The different colors observed in the ferric chloride tests indicate the formation of complexes with different properties depending on the nature of the complexes (Zewudie *et al.*, 2023). In this study, a biogenic synthesis of nanoparticles employing a bottom-up method has been carried out using phytochemicals extracted from *Lippia abyssinica* that might act as reducing and stabilizing agents in the process (Pandit *et al.*, 2022). The phytochemical composition of *Lippia abyssinica* leaf extract (LALE) was analyzed qualitatively using the standard procedure to determine the presence of some phytochemical-constituents in the leaf extract of the medicinal plant. The confirmatory tests, which involve color changes, precipitate formation, and others, were used to determine the presence or absence of useful bioactive ingredients such as alkaloids, tannins, flavonoids, flavones, terpenoids, phenols, and saponins in leaf extracts. The phytoconstituents' characteristics of the LALE are summarized in Table 3 and Appendix 1. Accordingly, the analysis carried out revealed that phenols, terpenoids, flavonoids, alkaloids, terpenoids, steroids, and saponins were present in the aqueous extract of the leaves of *Lippia abyssinica*. The results were in agreement with previously reported results (Agidew, 2022; Buli, 2015; Belachew *et al.*, 2022). The synthesis of the nanoparticles was significantly influenced by the presence of these bioactive components, which potentially serve to reduce, cap, and stabilize them, minimizing the chances of agglomeration.

Table 3. Preliminary phytochemical screening profile of the LALE

S.N	Metabolites	Test response	Methods
1	Phenol	+	<i>Ferric chloride test</i>
2	Flavonoids	+	<i>Alkaline reagent test</i>
3	Saponins	+	<i>Frothing test</i>
4	Tannins	+	<i>Ferric chloride test, Lead acetate test</i>
5	Terpenoids and steroid	+	<i>Salkowski test</i>
6	Alkaloids	+	<i>Mayer's reagent test</i>
+ Present			

4.4. Silver nanoparticle (AgNPs) synthesis

The syntheses of AgNPs were carried out under optimized conditions using the plant extract as a reducing and capping agent. On adding leaf extract, the colorless silver nitrate solution turned into a deep brown solution indicating the formation of AgNPs as shown in Figure 9. This is in agreement with previous studies on different plants (Hieu *et al.*, 2022; Mehata, 2021; Melkamu and Bitew, 2021). The presence of AgNPs was confirmed by the immediate change in color of the reaction mixture from colorless to brown after the addition of plant leaf extract. The immediate change in color of the reaction mixture is mainly due to the fact that the existence of localization SPR as result of free-moving electrons in AgNPs that interact with UV-vis radiation (Anandalakshmi *et al.*, 2016).

The mechanism of AgNPs synthesis could be explained by the electron-donating capacity of the plant extract's biomolecule content (polyphenol, alkaloid, saponin, tannin, terpenoids, and flavonoids) resulting in the reduction of Ag^+ ions to Ag metal. In addition, the synthesized AgNPs were stabilized by the bioactive compounds' functional groups like carbonyl or hydroxyl (alcohol) that keep them from agglomeration. For example, the hydroxyl groups in saponins interact with the Ag^+ ions to form AgNPs and provide stability for the newly formed nanoparticles (Purohit *et al.*, 2022). By interacting with the metallic ions through the extract's carbonyl functional groups, flavonoids release reactive hydrogen, which then changes the flavonoid's enol into keto form, resulting in the formation of Ag^0 (Shaikh *et al.*, 2021). The illustration in Appendix 2 shows some proposed bio-reduction mechanisms of AgNPs by phytochemicals and amino acids.

4.4.1. UV-Vis spectroscopy analysis of synthesized silver nanoparticles

UV-Vis spectroscopy was used to monitor the bio-reduction of silver ions to AgNPs. The size, shape, and production of the AgNPs in aqueous suspensions can be determined using UV-Vis spectroscopy (Wan Mat Khalir *et al.*, 2020). The absorbance of the as-synthesized nanoparticles was recorded in the scanning range of 200 – 800 nm in DMSO solution. Metallic nanoparticles have characteristic optical absorption spectra in the UV-Vis region. Electrons are limited to specific vibration modes by the particle's size and shape. The dispersions of

silver nanoparticles showed an intense band due to the SPR absorption. The surface of a metal is like plasma, having free electrons in the conduction band (Ershov *et al.*, 2021) and positively charged nuclei. SPR is a collective excitation of the electrons in the conduction band, near the surface of the nanoparticles. The band gap of the synthesized silver nanoparticle from Figure 10 was estimated to be about 2.45 eV, which is comparable to previously reported results (Joy *et al.*, 2021).

As shown in Figure 10, the UV-vis peaks of AgNO₃, LALE, and AgNP were observed at 275, 301, and 433 nm, respectively. The UV-vis spectrum of the LALE has a band of $n \rightarrow \pi^*$ transition of charge within CO, O-C=O, OH groups the phytochemical at 301 nm are observed that absorption often occurs in the range 250-300 nm due to electronic transitions or delocalization of electron (Akinpelu *et al.*, 2019). A UV-Vis spectrum of a solution of silver nitrate (AgNO₃) showing an absorption peak at around 275 nm is likely due to the electronic transitions involving the nitrate ions (NO₃⁻). The appearance of a typical SPR peak between 400 and 500 nm (Somee *et al.*, 2018) further supported the color change's indicating that AgNPs were formed. The absorbance band maxima of AgNP appeared in the range 400 - 450 nm due to the surface resonance of AgNPs (Meva *et al.*, 2016). This is also in good agreement with previously reported results using different plants, for instance, AgNP synthesis with *Solanum trilobatum* (430 nm), *Cinnamomum tamala* (416 nm)(Al Mashud *et al.*, 2022) and *Eichhornia Crassipes* (450 nm).

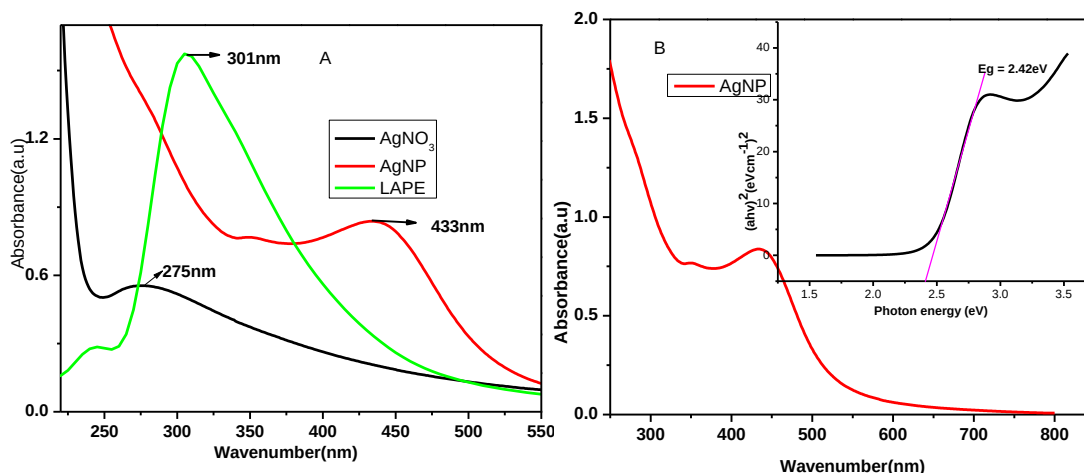


Figure 10. UV-Visible Spectra of (A) AgNO₃, LALE, and AgNP formed using LALE and (B) band gap of the **biogenic** AgNP.

4.4.2. Optimization parameters using UV-Vis spectroscopy for the synthesis of AgNPs

Optimization of various parameters including time, temperature, pH, the volume of the plant leave extract, and silver nitrate concentration were conducted to assess their influence on the biogenic of AgNPs. Accordingly, the plant leave extract, which was employed as a capping and reducing agent, helped to make it possible to observe the peak of the AgNP formed at a redshift (bathochromic) compared to the precursors AgNO_3 (Figure 11A).

4.4.2.1. Influence of AgNO_3 concentration on the biogenic synthesis of AgNPs

The optimization of silver ion concentration was done by taking 2, 4, 8, 12, and 16 mM from 50 mL aqueous solutions of silver nitrate. The changes in the reactions were monitored by UV-Vis spectroscopic scanning (Figure 11B). The production rate of the AgNPs increased with decreasing in AgNO_3 concentration. The UV-Vis spectral data revealed that the optimum concentration of AgNO_3 was 2 mM, above which the product quantity started to decline, hence at a higher concentrations of AgNO_3 , the absorbance was found to decrease and shift to blue (from 416 to 397 nm), in agreement with a previous study (Htwe *et al.*, 2019). The experimental design that concentrated on concentrations above the previously reported optimum (2 mM) makes sense for a number of reasons in order to maximize the production yield of silver nanoparticles (AgNPs). This work positions the experiment to determine a concentration that may maximize the yield of AgNPs under particular synthesis conditions by concentrating on silver nitrate concentrations above the previously reported optimum. It This is promising to give priority to concentrations that are most likely to produce larger yields, even though testing below 2 mM may reveal information on the bottom limit of Ag ion availability.

4.4.2.2. Effect of volume ratio of AgNO_3 to LALE

Figure 11C shows the absorption spectra of AgNPs as the function of the volume of the plant leave extract as (2, 4, 6, 8, and 10 mL) extract. With the increase in the concentration of LALE in the reaction mixture (1:5), the peak intensity increases abruptly, indicating an enhancement

in the production of AgNPs (Ali *et al.*, 2016). Additionally, as the plant leave extract volume increased, the absorption spectrum slightly shifted toward longer wavelength region (from 414 to 434 nm), indicating that AgNP size had increased as the spectra shifted to red (Ali *et al.*, 2016).

4.4.2.3. Effect of temperature on the synthesis of AgNPs

To optimize the reaction temperature, each reaction was run independently for two hours at various temperatures (10, 20, 50, 80, and 100 °C) is shown in Figure 11D. At a temperature of 80 °C, nanoparticles of good stability and high yield were obtained. As a result, in this study, the optimum temperature for AgNP synthesis was set at 80 °C. In the optimization of reaction temperature for AgNPs synthesis, temperatures lower than 80 °C (e.g., 10 °C, 20 °C, and 50 °C) likely resulted in insufficient energy for the reduction of silver ions (Ag^+) by the plant extract or reducing agent, leading to slower reaction kinetics, lower nanoparticle yield, and possible formation of less stable or larger-sized particles. At these lower temperatures, the reduced rate of reduction may also prevent the uniform growth of nanoparticles, resulting in a less controlled size and shape. On the other hand, at temperatures higher than 80 °C (e.g., 100°C), the rapid reduction rate could lead to uncontrolled particle nucleation, causing nanoparticles to aggregate or form non-uniform shapes, thereby reducing stability. Excessive heating may also degrade the stabilizing phytochemicals or capping agents in the plant extract, leading to nanoparticle instability and poor quality. Therefore, 80 °C is identified as the optimal balance where sufficient energy is provided for controlled reduction, resulting in nanoparticles with good stability and high yield (Asif *et al.*, 2022; Jiang *et al.*, 2011).

4.4.2.4. The reaction time effect on the synthesis of the AgNPs.

The reactions for the phyto-mediated synthesis of nanoparticles were carried out at various time intervals ranging from 0 to 4 hours as shown in Figure 11A. The absorption spectra were scanned after 0 min, 30 min, 1 h, 2 h and 4 h. At 2 hours, the maximum absorbance was recorded. The results showed that lower absorption peaks at 0 min, 30 min, and 1 h indicate lower AgNP yields, reaction rates, and stability. With increasing time, the absorbance has

increased, which suggests that the formation of AgNPs has improved. As a result, 2 h was considered as the ideal time for AgNP synthesis, which served as the foundation for additional optimization.

4.4.2.5. The effect of pH for the green synthesis of AgNPs

The effect of pH on the biogenic of silver nanoparticles was studied at pH values of 2.99, 5.58, 6.99, 8.34, 10.34, and 12.01 keeping the time at 2 h using UV-Vs spectroscopy (Figure 11E).

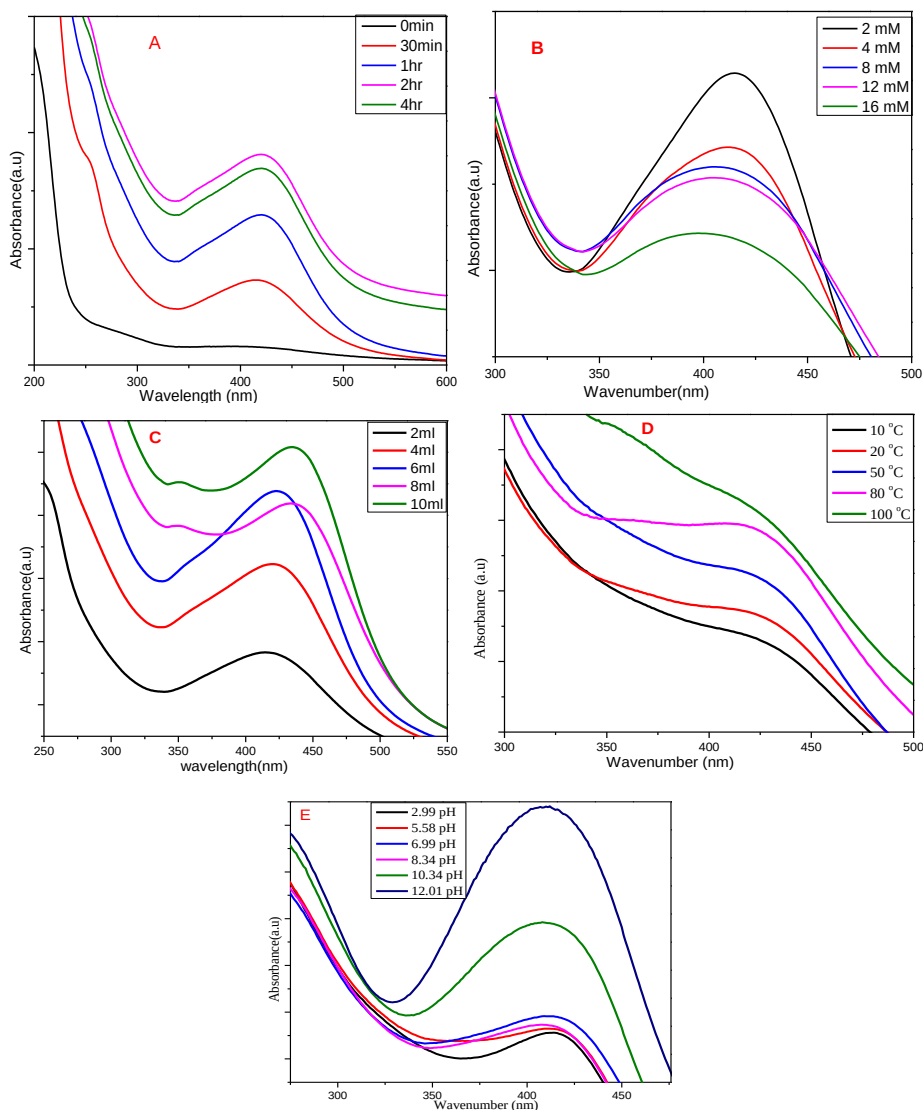


Figure 11. Surface Plasmon resonance spectra of **biogenic** AgNPs recorded as a function of (A) time, (B) concentration of AgNO_3 solution, (C) concentration of extract, (D) temperature and (E) pH effect.

The biogenic of silver nanoparticles at pH 2.99, 5.58, 6.99, and 8.34 was slower than at pH 10.34, and 12.01, as depicted in the absorption spectra. However, at pH 12.01, high absorption spectra were recorded, indicating that the formation of AgNPs was successful and the product was stable, resulting in high yield. Thus, the synthesis of the AgNPs was carried out under optimized conditions of 2 mM of AgNO₃, a 1:5 metal salt to LALE concentration ratio, a temperature of 80 °C, with a reaction time of 2 h at pH of 12.01. These conditions were considered suitable for the biogenic of the AgNPs.

4.4.3. Functionalization of the biogenic AgNPs by amino acids

The use of NPs for drug design systems (DDS) can improve the side effects observed in existing pharmacological treatments through the functionalization of NPs for specific targets (Patra *et al.*, 2018). The physicochemical features of NPs, such as size, shape, surface charge, surface properties, and conjugation with specific biological molecules, are used to improve the delivery of drugs to the target tissue in two diverse ways: passive and active targeting (Zein *et al.*, 2020). In this study, we had functionalized the biogenic AgNP with histidine and phenylalanine amino acids. The optimization was done for 0.01 M histidine and phenylalanine considering volume of 2, 4, 8 and 10 mL for both amino acids. The UV-visible spectra data were recorded for each volume at 2 h, 1:5 volume ratio of AgNO₃ to LALE, 80 °C, and pH 12.01. As shown in Figure 12 A and B, 4 mL and 6 mL of histidine and phenylalanine, respectively, showed high absorption peaks, which confirm the formation of stable functionalized AgNPs. As shown in Figure 12C, the biogenic AgNPs functionalized with amino acid absorbance peak shifted to blue indicated that it increases surface area and decreases the size of the biogenic AgNPs. The shift in UV-vis absorbance from 414 nm (for AgNPs synthesized using plant leaf extract alone) to 401 nm (when amino acid is added) suggests that the amino acid plays a role in reducing the size of the nanoparticles. This blue shift typically indicates smaller particle size, as smaller nanoparticles exhibit SPR peaks at lower wavelengths. The plant extract acts as an effective capping agent initially, stabilizing the silver nanoparticles and preventing excessive growth, as reflected in the peak at 414 nm. However, the introduction of the amino acid provides a secondary capping layer, which

further stabilizes the particles and restricts their growth. This additional capping results in smaller, uniform nanoparticles, as evidenced by the shift to 401 nm the wavelength of silver nanoparticle. The plant extract shows good capping efficiency, but the presence of the amino acid enhances the nanoparticle stability and size control, leading to the formation of smaller AgNPs (Allahverdiyev *et al.*, 2011; Shankar and Rhim, 2015)

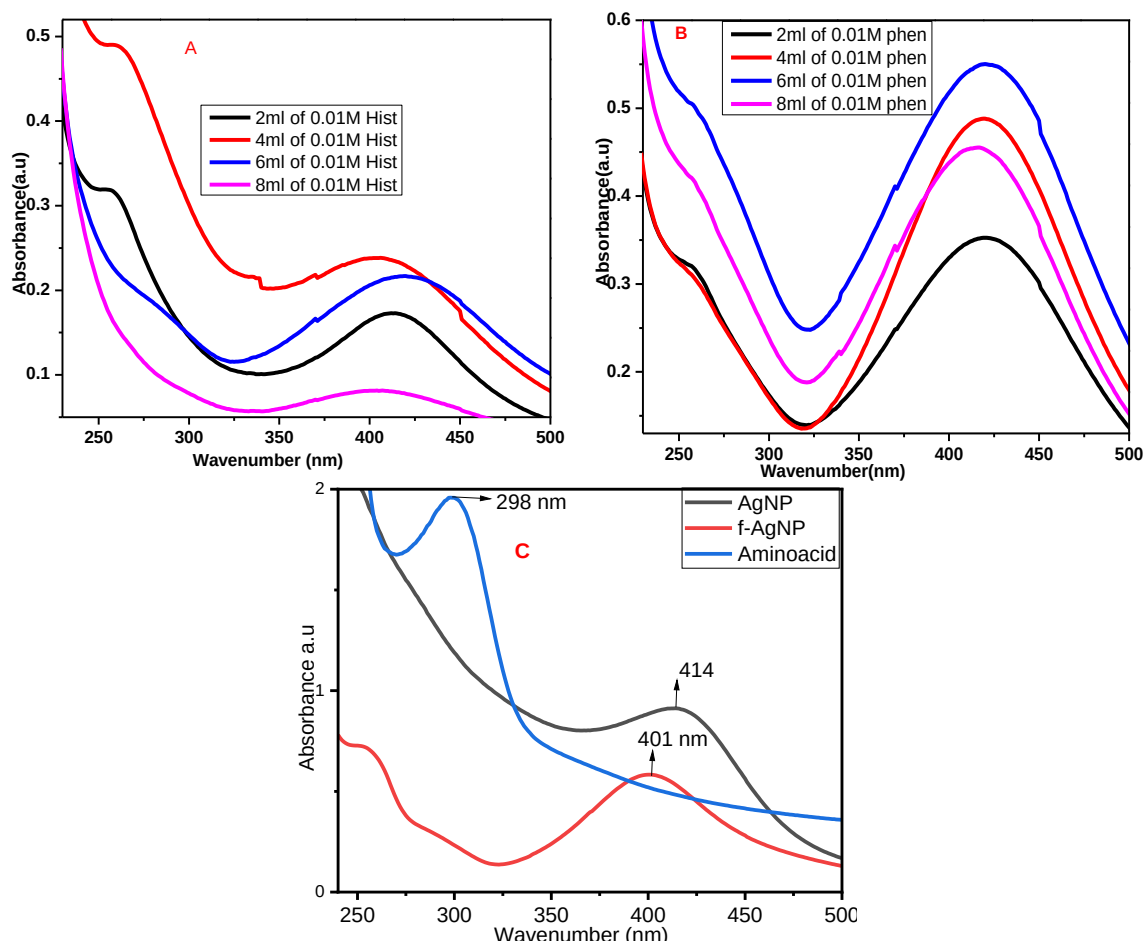


Figure 12. Optimization of the volume of 0.01M of (A) Histidine and (B) Phenylalanine for functionalization of the green AgNP synthesis and (C) Comparison of f-AgNP with others.

4.5. The UV-Vis spectra of imine-quinoline ligand (L), Zn(II) and Cu(II) complexes

The electronic spectra and computed with TD-DFT of the imine quinoline ligands (L) and its Zn(II) and Cu(II) complexes are given in Table 4 and Figure 13 below and Appendix 4-5. The ligand showed three major band at 259 nm, 308 nm and 394 nm. The band 259 nm is due to

the $\pi \rightarrow \pi^*$ transition of ligand centered transition (LCT) of benzene. The band appearing at 308 nm is as a result of $n \rightarrow \pi^*$ transition non-bonding electrons present on the nitrogen of the azomethane group (-HC=N). The band at 394 nm is due to $n \rightarrow \pi^*$ transition of the phenolic group. (Smolková *et al.*, 2023; Eichhorn *et al.*, 2023). Zn(II) complexes (ZnL) displayed absorption bands at 259 nm, 312 nm, and 394 nm. The bands at 259 nm and 312 nm were due to intra-ligand transition (ILT) ($\pi \rightarrow \pi^*$) and the band at 394 nm was assignable to the metal to ligand charge transfer (MLCT) ($n \rightarrow \pi^*$) transition. An octahedral geometry is proposed for this complexes based on its analytical, conductance and spectral data. This is further supported by its diamagnetic nature and absence of d-d band, due to its complete d^{10} electronic configuration. Similarly, ZnL₂ displayed three-absorption band at 261 nm, 308 nm and 394 nm. The bands at 261 nm and 308 nm were due to intra-ligand transition (ILT) ($\pi \rightarrow \pi^*$) and the band at 394 nm was assignable to the metal to ligand charge transfer (MLCT) ($n \rightarrow \pi^*$) transition. MLCT band position around 250-350 nm for systems where the ligand has high-energy π^* orbitals or the metal has low-energy d orbitals like Zn(II) complexes (Eichhorn *et al.*, 2023; Smolková *et al.*, 2023). CuL displayed four absorption bands at 262 nm, 322 nm, 386 nm, and 588nm. These assignments suggest a combination of both ligand-centered transitions and metal-related transitions (MLCT and d-d) in the electronic absorption spectrum of the Cu(II)-imine quinoline complex (Abu-Dief *et al.*, 2024). At 262 nm absorbance band is likely due to the ligand-centered ($\pi \rightarrow \pi^*$) transition within the imine quinoline ligand. Such transitions are common for aromatic ligands like quinoline and occur at shorter wavelengths (higher energies). 322 nm absorption band may still be the within ligand, possibly another $\pi \rightarrow \pi^*$ transition or an $n \rightarrow \pi^*$ transition. 386 nm absorption band is expected due to a metal-to-ligand charge transfer (MLCT) transition, where an electron from a d-orbital of the Cu(II) metal center is transferred to a π^* orbital of the ligand. 588 nm absorption band in the visible region is most likely due to a d-d transition within the Cu(II) centered that is responsible for the complex's colour. The observed shifts in all bands and the formation new bands in complexes confirmed that the reaction had occurred between Cu(II) and the ligand.

Transitions resulting from the metal ion's d orbital and intra-ligand transitions are seen at 592/580 nm (experiment/TD-DFT) for CuL. The result allows one to ascertain the degree of fission associated with a certain geometry because of extracted transitions such d $\rightarrow$ d

electronic transition, MLCT, and intra-ligand transitions (Kumar *et al.*, 2011). Transitions like $n \rightarrow \pi^*$ and $\pi \rightarrow \pi^*$ in coordinating ligands have been seen in deep ultraviolet areas. Jahn Teller's distortion causes the Cu(II) complexes' UV-Vis spectra to have deformed octahedral geometry. Moreover, transitions at 401/399 nm is linked to $d \rightarrow d$ ($t_{2g} \rightarrow e_g$) transitions (Abu-Dief *et al.*, 2024; Abbas *et al.*, 2024).

Table 4 Assignment of the important $\lambda_{\max}$ (nm) of the imine-quinoline ligand and its Cu(II) and Zn(II) complexes

Complex	Color	Absorbance (nm) experimental	Absorbance (nm) Calculated	Transition	Assignment	Geometry
L	Yellow	259	223, 259	$\pi \rightarrow \pi^*$	LCT	
		308	298	$\pi \rightarrow \pi^*$		
		394	372	$n \rightarrow \pi^*$		
[ZnL(H₂O)Cl₂]	Light	259	223, 259	$\pi \rightarrow \pi^*$,	ILT	Octahedral
C₂₂H₂₀Cl₃N₃O₃SZ	yellow	312	299	$\pi \rightarrow \pi^*$,		
n		390	383, 450	$n \rightarrow \pi^*$	MLCT	
[ZnL₂Cl₂]	Light	261	261,	$\pi \rightarrow \pi^*$,	ILT	Octahedral
C₄₄H₃₂Cl₄N₆O₂S₂Z	yellow	308	287,	$\pi \rightarrow \pi^*$,		
n		394	397	$n \rightarrow \pi^*$	MLCT	
[CuL(H₂O)₂Cl₂]	Brick	262,	270,	$\pi \rightarrow \pi^*$,	ILT	Octahedral
C₂₂H₂₀Cl₃CuN₃O₃	red	322,	318,	$\pi \rightarrow \pi^*$,		
S		386,	399	$n \rightarrow \pi^*$,	MLCT	
		588	461	$d-d$	$^2T_{2g} \rightarrow ^2E_g$	
[CuL₂Cl₂]	Blue	262,	255,	$\pi \rightarrow \pi^*$,	ILT	Octahedral
C₄₄H₃₂Cl₄CuN₆O₂		306,	333,	$\pi \rightarrow \pi^*$,		
S₂		386,	418,	$n \rightarrow \pi^*$,	MLCT	
		540	540	$d-d$	$^2T_{2g} \rightarrow ^2E_g$	

The intramolecular charge transfer bands of $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ transitions of the azomethine group $C=N$ is responsible for the absorption band at 265/270 nm. These values also coincide with those of related Schiff-base compounds (Alam et al., 2023; Manvatkar *et al.*, 2023). Similarly, CuL_2 displayed four-absorption band at 262 nm, 306 nm, 386 nm, and 540 nm. The band at 540 nm was as result of d-d spin allowed transition for ${}^2T_{2g} \rightarrow {}^2E_g$. This transition is suggestive of octahedral geometry around $Cu(II)$ ion. Overall, the experimental results were found to be in a good agreement with those from the TD-DFT calculations as shown Table 4 and Figure 14. These results further validate the suggested structures the complexes to be octahedral.

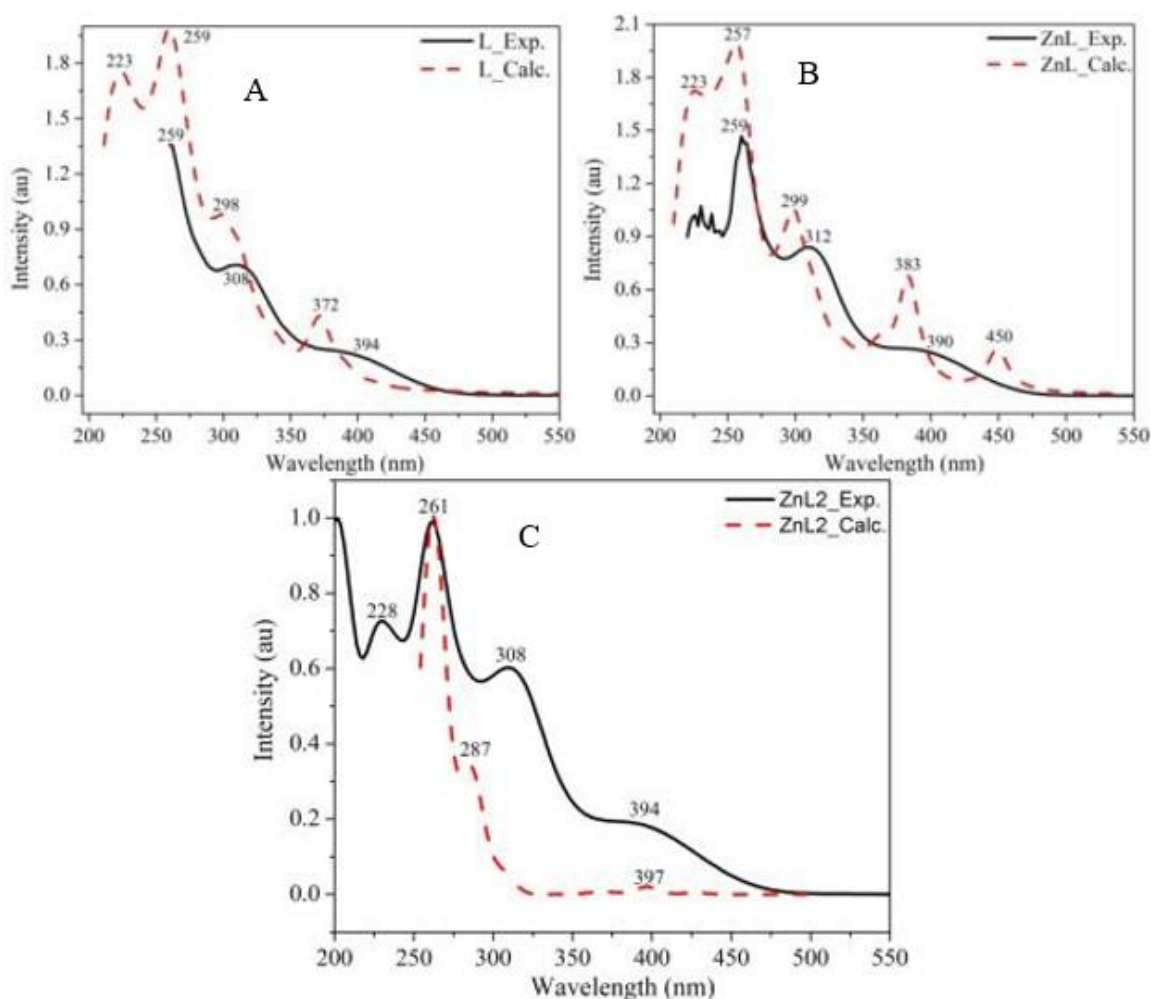


Figure 13. UV-vis spectrum and TD-DFT of the (A) ligand (L), (B) ZnL and (C) ZnL₂

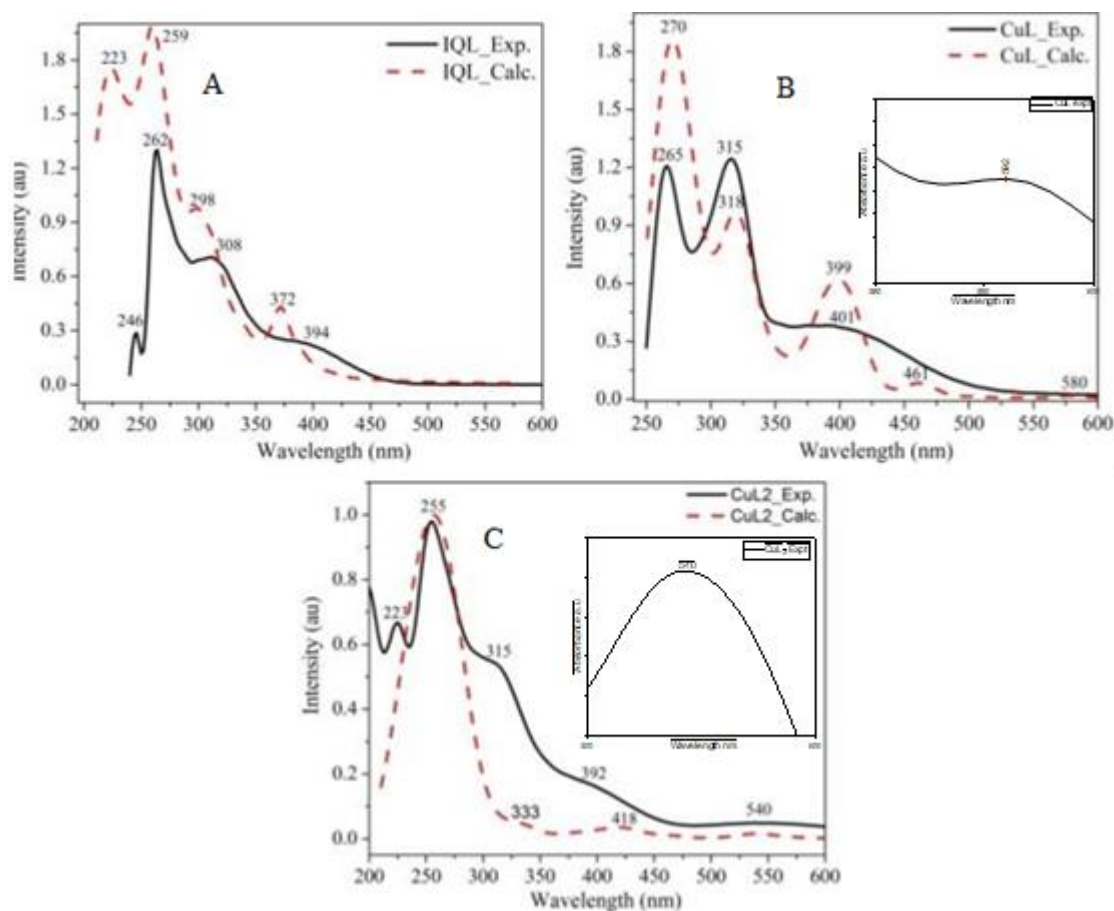


Figure 14. UV-vis spectrum and TD-DFT of the (A) ligand (L), (B) CuL and (C) CuL₂

4.6. The UV-Vis spectra of decorated AgNP with ligand, Zn(II) and Cu(II) complexes

The electronic absorption spectral data are presented in the Table 5 and Figure 15. The silver nanoparticle decorated with the ligand (AgNP@L) showed three major bands at 238 nm, 300 nm, and 384 nm. The AgNP@L exhibits an intra-ligand charge transfer (CT) peak at 384 nm. The decorated and undecorated ligands and their metal complexes' electronic absorption spectra show almost the same wavelengths with a few band changes was observed from the spectra as seen in the UV-Vis spectra of imine-quinoline ligand (L), Zn(II) and Cu(II) complexes sections and Figure 16. These changes might be caused by variations in the target electron density. In addition to these absorption peaks, decorated metal complexes produce peaks in the high-wavelength areas.

Table 5. The UV-Vis maximum wavelength of the imine quinoline ligand and Cu(II) and Zn(II) complex decorated AgNP

Complex	Color	Absorbance (nm) experimental	Transition
AgNP@L	Yellow	238	$\pi \rightarrow \pi^*$
		300	$\pi \rightarrow \pi^*$
		384	$n \rightarrow \pi^*$
AgNP@ZnL	Light yellow	222, 240	$\pi \rightarrow \pi^*$
		302	$\pi \rightarrow \pi^*$
		396	$n \rightarrow \pi^*$
AgNP@ZnL₂	Light yellow	214, 238	$\pi \rightarrow \pi^*$
		296	$\pi \rightarrow \pi^*$
		390	$n \rightarrow \pi^*$
AgNP@CuL	Brick red	238, 300	$\pi \rightarrow \pi^*$, $\pi \rightarrow \pi^*$
		390	$n \rightarrow \pi^*$
		556	d-d
AgNP@CuL₂	Blue	238, 296	$\pi \rightarrow \pi^*$, $\pi \rightarrow \pi^*$
		382	$n \rightarrow \pi^*$
		556	d-d

As shown in both Figures 16 and Table 5 the UV-vis spectra were recorded for the AgNP decorated with ligand, and the peaks are 238, 300, and 384 nm. There are absorbance shifts between the imine-quinoline ligand (L) and AgNP@L. The silver nanoparticle decorated with ZnL (AgNP@ZnL) showed three major bands at 222, 240, 302, and 396 nm that there is an absorbance peak shift between ZnL complex and AgNP@ZnL. The absorbance peaks of AgNP-his decorated with ZnL₂ (AgNP@ZnL₂) were recorded at 238, 296, and 390 nm, and a change in absorbance peaks between AgNP@ZnL₂ and ZnL₂ was observed. Similarly, the UV-vis spectra of AgNP decorated CuL (AgNP@CuL) and AgNP decorated with CuL₂ (AgNP@CuL₂) were recorded as (238, 300, 390, 556) nm and (238, 296, 382, 556) nm, respectively.

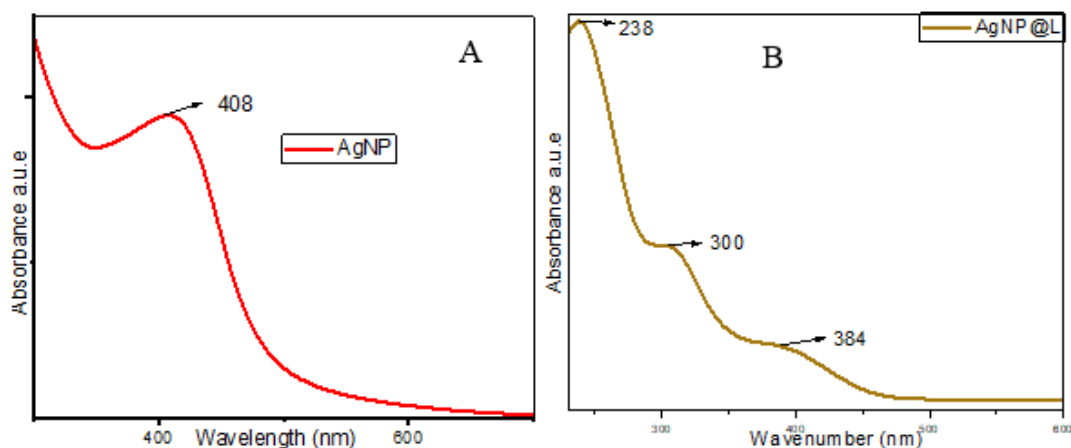


Figure 15: The UV-Vis spectra of A) AgNP and B) decorated Ligand (AgNP@L)

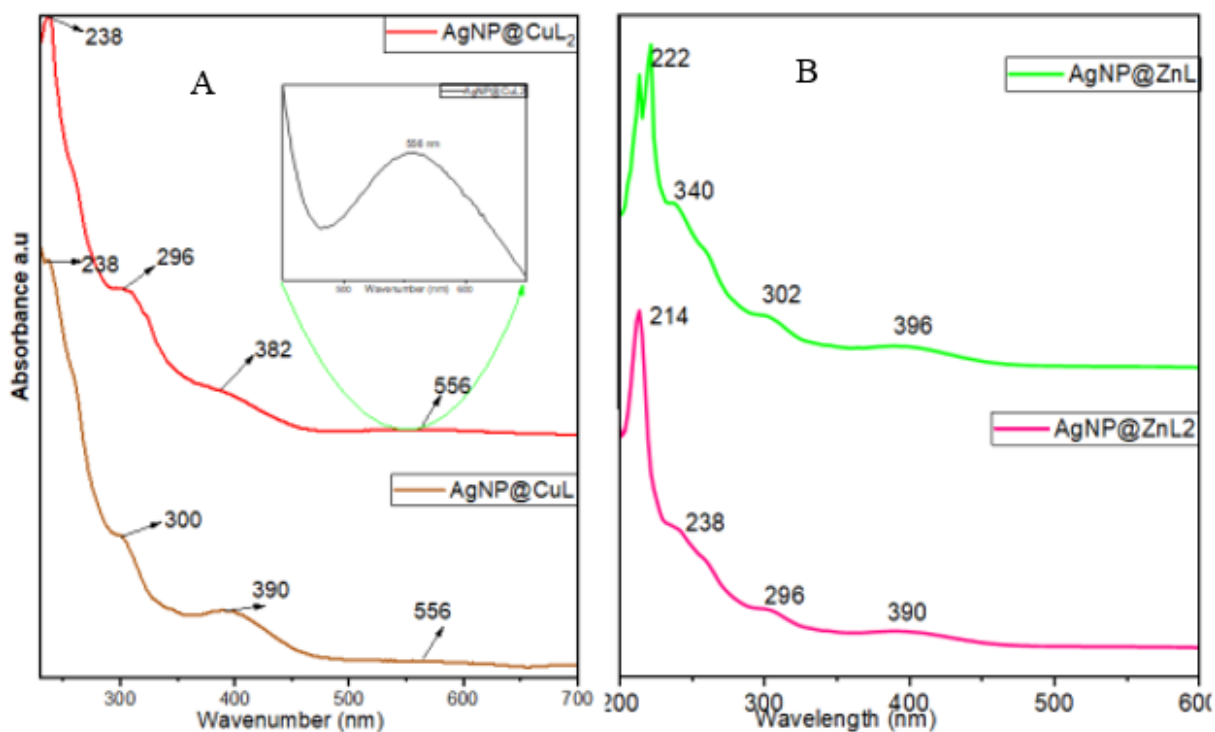


Figure 16: UV-vis spectrum a of the (A) AgNP@Cu(II) complexes and (B) AgNP@Zn(II) complexes.

These transitions involve both the ligand and the metal center, with ligand-centered transitions occurring at higher energy (shorter wavelengths in the UV region) and metal-involved transitions (MLCT) at lower energy (longer wavelengths, including the visible region). There is a shift in absorbance between the Cu(II) complexes and the AgNPs decorated with the

Cu(II) complexes. The change in absorbance shift indicated the influence of AgNPs, and the changes are due to decoration. As shown in Figure 15 and 16, the absorption band of the AgNP-his disappeared when decorated with the ligand and its Cu(II) and Zn(II) complexes. The disappearance of the AgNP absorption band, when decorated with the ligand and its Cu(II) and Zn(II) complexes, could result from SPR damping, changes in the local dielectric environment, aggregation of the AgNPs, or the formation of a new hybrid material with altered optical properties. Additionally, oxidation or surface passivation of the AgNPs due to the complexation or adsorption of these metal-ligand species on the surface of AgNPs can lead to surface modification that suppresses the characteristic SPR response of AgNPs (Matlou and Nyokong, 2020b).

The absence of the characteristic absorbance band of AgNPs in the decorated with imine quinoline ligands and their Cu(II) or Zn(II) complexes can be attributed to strong interactions between the AgNPs and the metal complexes. These interactions likely result in the quenching or masking of the AgNP surface plasmon resonance (SPR) band, typically observed between 400–450 nm. The coordination of the metal complexes to the surface of the AgNPs could alter their electronic environment, disrupting the nanoparticles' plasmonic properties. This leads to the suppression of the AgNP absorbance band in the composite spectra. Additionally, the strong absorption of the ligands and their metal complexes may dominate the spectrum, overwhelming the weaker SPR signal of AgNPs and making it difficult to detect.

Another factor could be surface modification of the AgNPs by the ligand or metal complexes, which may shift or broaden the SPR band beyond recognition. The binding of these ligands and complexes to the AgNP surface can alter the nanoparticles optical properties, potentially causing aggregation or size changes that further contribute to the disappearance of the characteristic SPR peak. Furthermore, the formation of a new composite material may create a structure with distinct optical properties, where the ligand-metal charge transfer transitions of the complexes dominate the absorbance spectrum, leaving the AgNPs plasmonic effects obscured. As a result, the observed spectra primarily reflect the optical characteristics of the ligands and their Cu(II) and Zn(II) complexes.

4.7. Nuclear magnetic resonance (^1H and ^{13}C NMR) analysis of the ligand and the zinc complexes

The proton NMR spectra of the ligand and zinc (II) complexes obtained in DMSO- d_6 , are displayed in (Table 5 and Appendix 6-11). The ^1H and ^{13}C NMR spectra of the imine quinoline ligand (L) showed a singlet peak at $\delta = 8.90$ ppm corresponding to the azomethine proton ($-\text{N}=\text{CH}-$) (Wang *et al.*, 2022) an indication that the Schiff base was formed during condensation reaction. Observation of a peak at $\delta = 151.18$ ppm in the ^{13}C NMR spectrum was further proof that the ligand was successfully synthesized (Wang *et al.*, 2022). The azomethine proton peaks of imine quinoline ligands (L) shifted up field in the ^1H NMR spectra of the ZnL and ZnL₂ complexes ($\delta = 8.88$ and 8.89 ppm respectively). The up field shifting of azomethine proton in ZnL and ZnL₂ complexes was attributed to the discharging of electronic cloud towards the Zn(II) ions indicating coordination through the azomethine nitrogen to the metallic ion. The expected ^1H NMR peak, at $\delta = 9.30$ ppm, assignable to the phenolic proton in the free ligand (Sayed *et al.*, 2023). In ZnL and ZnL₂ complexes, this signal changes to $\delta = 9.10$ and 9.19 ppm, respectively, showing complexation due to the bonding of ligands with the metal centers (Wang *et al.*, 2022). These changes confirm that the imine chelation with metal attributable to the bond formation between Zn(II) metal ion and nitrogen of imine quinoline ligand. A singlet approximately at $\delta = 3.47$ ppm in the newly emerged chemical shift peak indicates the freshly formed metal-water combination in the ZnL complex (Rimsza *et al.*, 2019; Abdoon *et al.*, 2022). The peaks shift to higher values depicts that the nitrogen atom in the moiety has released its electrons to the zinc (II) metal. The steric effects and zinc (II) complex geometry may be the cause for the peaks downward shifting. Protons in the phenyl ring moved to up field during complexation, supporting the creation of Zn(II) complexes, and proves that the singlet, doublet, and multiplet seen around $\delta 8.11$ – 6.93 ppm correspond to these ^1H (proton) (Abdoon *et al.*, 2022).

Furthermore, the synthesis of Zn(II) complex and the imine quinoline ligand were supported by the ^{13}C NMR spectrum data (Figure S4 & S5). The carbon atom of the imine $-\text{HC}=\text{N}-$ group was at $\delta = 151.18$ ppm (Hernández *et al.*, 2020) has shifted in the Zn(II) complexes to $\delta = 149.24$ ppm and 151.16 ppm of ZnL and ZnL₂, respectively. This indicate that the nitrogen

atom of the imine-C=N-group has bonded to the metal center, shifting the N-atom's electron density in the direction of the zinc (II) metal ions. The carbon atom carrying a hydroxyl group, or phenol (=C-OH), in the ligand that makes up the imine quinoline ligand was found to have a significant peak at δ 155.92 ppm in the Zn(II) complex spectra, which displays the hydroxyl group by coordinating via metal ion (Khalil *et al.*, 2021). The shift observed around δ 155.88–155.89 ppm implies the successful complexation of the ligand with the Zn(II) metal ions. The aromatic carbon's chemical shift signals are often assigned to quinoline derivative ligands in the range of δ 148.52–111.77 ppm (Gunawan *et al.*, 2021). These ligand signals were shifted due to the complex formation with metals as indicated in the ^{13}C and ^1H NMR spectrum data.

Table 6. ^1H and ^{13}C NMR chemical shift data of imine quinoline ligand (L) and its Zn(II) complexes

Chemical shifts (δ) ppm			
L	ZnL	ZnL ₂	Assignments
^1H NMR			
9.20 (s)	9.10(s)	9.19(s)	Phenolic OH
8.90 (S)	8.89 (s)	8.88 (s)	Azomethine (imine) proton
5.40 (s)	5.39(s)	5.40(s)	Amine proton
7. 81-8.4	8.64 – 8.08(m)	8.38 – 7.33(m)	Pyridine ring proton
6.61-7.31	7.32 – 7.27 (m)	7.32 – 7.27(m)	Ar-H proton
	3.45		M-O-H ₂
^{13}C NMR			
151.18	151.16	151.16	-HC=N
155.92	155.89	155.88	C-OH
131.95	131.93	131.93	C-Cl
133.77	133.77	133.77	C-NH ₂

The fact that, the phenol group of the imine quinoline ligand forms a complex with Zn(II) in an octahedral geometry (with additional ligands like Cl^- and H_2O) without deprotonation, as evidenced by a small shift in the ^1H NMR spectrum from δ = 9.3 ppm to 9.10 ppm (ZnL), and to 9.19 ppm (ZnL₂), suggests that the phenolic proton remains intact during complexation. The

phenol group remains protonated when the imine quinoline ligand complexes with Zn(II) because the metal's octahedral coordination is sufficiently satisfied by the quinoline nitrogen, imine nitrogen, chloride ions (Cl^-), and water molecules (H_2O). The phenol oxygen likely interacts weakly with the Zn(II) center, possibly via hydrogen bonding, without requiring deprotonation. This is evidenced by the small downfield shift in the ^1H NMR spectrum from $\delta = 9.3$ ppm to 9.10 ppm or 9.19 ppm, indicating a slight electronic interaction.

NMR Spectral data of the imine quinoline ligand (L) and its Zn(II) complexes

Imine quinoline ligand (L)

^1H NMR (400 MHz, DMSO) 9.20 (s, 1H), 8.90 (s, 1H), 8.39 (d, $J = 8.8$ Hz, 1H), 8.16 (d, $J = 2.0$ Hz, 1H), 7.80 (dd, $J = 8.8, 2.1$ Hz, 1H), 7.30 (ddd, $J = 12.3, 7.8, 1.6$ Hz, 2H), 7.26 – 7.12 (m, 3H), 6.82 (dd, $J = 8.2, 1.4$ Hz, 1H), 6.74 – 6.57 (m, 2H), 5.40 (s, 2H).

^{13}C NMR- (100 MHz, DMSO) 155.92, 151.18, 148.53, 148.06, 138.69, 137.76, 137.57, 133.77, 133.37, 131.95, 131.61, 129.37, 129.24, 127.98, 127.89, 127.11, 126.53, 125.96, 125.90, 118.79, 117.36, 117.23, 115.29, 111.77.

ZnL:

^1H NMR (400 MHz, DMSO) δ 9.10 (dd, $J = 64.2, 4.4$ Hz, 1H), 8.88 (s, 1H), 8.64 – 8.08 (m, 2H), 8.08 – 7.51 (m, 2H), 7.30 (dd, $J = 14.2, 7.9$ Hz, 2H), 7.18 (ddd, $J = 24.6, 16.4, 8.7$ Hz, 2H), 7.09 – 6.96 (m, 2H), 6.85 – 6.47 (m, 2H), 5.39 (s, 2H), 3.47 (s, 4H).

^{13}C NMR (100 MHz, DMSO) δ 155.89, 151.16 (d, $J = 5.2$ Hz), 150.90, 149.24, 148.80, 148.51, 148.04, 141.73, 139.08, 138.67, 137.75, 137.56, 133.76, 133.34, 132.52, 131.92, 131.62, 129.42 (d, $J = 7.2$ Hz), 129.23, 127.92 (d, $J = 7.0$ Hz), 127.13 (d, $J = 8.1$ Hz), 126.56, 125.92, 125.55, 118.78, 117.34 (d, $J = 10.2$ Hz), 116.34, 115.26 (d, $J = 5.5$ Hz), 111.80.

ZnL₂:

^1H NMR (400 MHz, DMSO) δ 9.19 (s, 0H), 8.89 (s, 0H), 8.38 (d, $J = 8.9$ Hz, 0H), 8.34 (d, $J = 8.9$ Hz, 0H), 8.16 (s, 0H), 8.14 (s, 0H), 7.81 (t, $J = 10.0$ Hz, 0H), 7.35 (s, 0H), 7.33 (s, 0H), 7.32 – 7.27 (m, 0H), 7.21 (dd, $J = 16.1, 8.0$ Hz, 1H), 7.16 – 7.07 (m, 0H), 7.03 (s, 0H), 7.01 – 6.96 (m, 0H), 6.82 (d, $J = 8.1$ Hz, 0H), 6.71 (s, 0H), 6.69 (s, 0H), 6.67 – 6.57 (m, 1H), 5.40 (s, 1H),

^{13}C NMR (100 MHz, DMSO) δ = 155.88, 151.16 (d, J = 2.3 Hz), 149.33 – 147.95 (m), 141.81 – 137.48 (m), 133.77 (d), 133.36 (d), 132.52 (d), 131.93 (d), 131.61 (d), 129.42 (d, J = 8.3 Hz), 129.23 (d), 127.92 (d, J = 7.6 Hz), 127.14 (d, J = 8.2 Hz), 126.54 (d), 126.10 – 125.48 (m), 119.16 (d, J = 77.1 Hz), 117.31 (d, J = 11.4 Hz), 116.33 (d), 115.24 (d, J = 7.0 Hz), 111.78 (d).

4.8. Fourier Transform Infrared (FT-IR) Spectroscopy analysis

4.8.1. FTIR Spectroscopy analysis of the AgNPs

The FT-IR analysis was carried out to obtain information about functional groups present in the AgNPs and understand the transformation of functional groups due to the reducing process. FTIR data were collected between 4000 and 400 cm^{-1} (Karthik *et al.*, 2020). The FT-IR spectra of the plant leave extract, AgNPs, and histidine and phenylalanine functionalized AgNPs were presented in Figure 17(A-D) as follows.

The spectrum of LALE shows absorbance peaks at 3850, - 3260 cm^{-1} (O-H stretching) (Oves *et al.* 2022); 2673, 2295, 2087, 1902, 1628 (C=O, flavonoids), and 1369, 1068, 941, 700 cm^{-1} (flavonoids) (Gupta *et al.*, 2022). The spectrum of non-functionalized AgNP showed absorbance peaks at 3376, 2417, 2282, 1653, 1395, 960, 868, 785, 682, 613, 573 cm^{-1} . On the other hand, the spectrum of functionalized AgNP-his showed absorbance peaks at 3846, 3718, 3491, 3308, 2785, 2320, 2193, 1680, 1475, 1384, 1286, 1169, 988, 910, 850, 771, 731, 659, 575 cm^{-1} , whereas functionalized AgNP-phen showed absorbance peaks at 3873, 3441, 2432, 2280, 1647, 1408, 1292, 954, 840, 615, 567, 447 cm^{-1} . The spectrum data showed the presence of the biomolecules, and there are some peaks shifting between LALE and the synthesized nanoparticles, indicating that LALE served as a capping agent and stabilizer for the synthesis of AgNPs. When compared to the FTIR spectrum of LALE, significant shifts for AgNPs were observed in the regions 1700-1500 cm^{-1} and 3600-3100 cm^{-1} , indicating the presence of carbonyl, hydroxyl, and amino groups at their surface which served to reduce Ag^+ to AgNPs because of their oxidation-reduction potentials. New peaks in a region 600-400 cm^{-1} were also observed due to the formation of AgNP new bonds with oxygen of carbonyl or carboxyl functional groups as a result of the presence of the biomolecules (Shahzadi *et al.*, 2022).

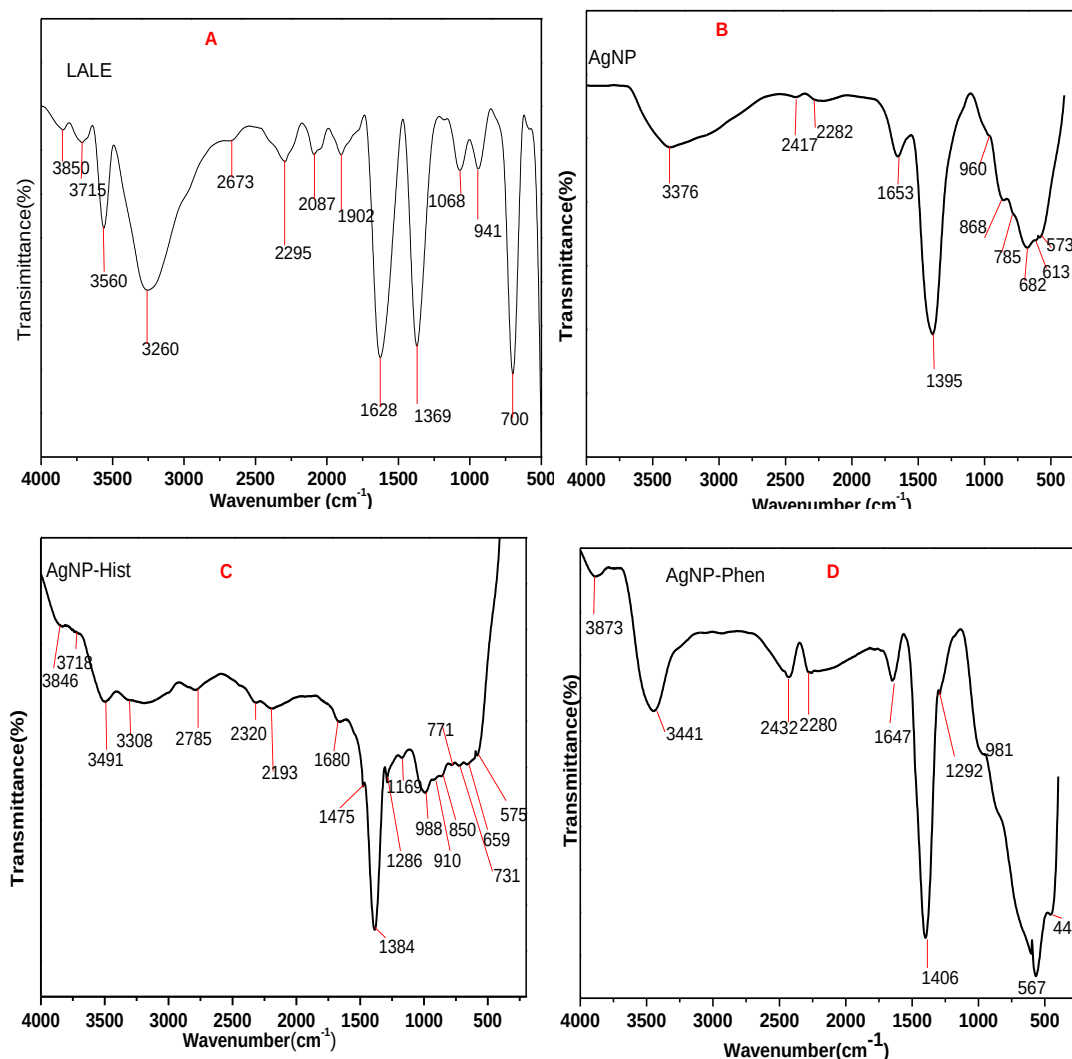


Figure 17. FT-IR spectra of *Lippia abyssinica* leaf extract (LALE (A)), biogenic AgNP (B), biogenic AgNP functionalized Histidine amino acid (AgNP-Hist (C)) and biogenic of AgNP functionalized Phenylalanine amino acid (AgNP-Phen (D)).

4.8.2. FTIR spectral analysis of the ligand (L) and its Zn(II) and Cu(II) complexes

The FT-IR results of the imine quinoline ligand (L) and its Cu(II) and Zn(II) metal complexes together with their spectra data are presented in Table 7 and Figure 18-21. The FTIR table provided presents the vibrational frequencies for a ligand (L) and its complexes with Cu(II) and Zn(II), labeled as CuL, CuL₂, ZnL, and ZnL₂. The key functional groups under investigation include amine (NH₂), imine (=NH), carbon-oxygen (C-O), carbon-nitrogen (C=N), hydroxyl (OH), water (H₂O), metal-oxygen (M-O), metal-nitrogen (M-N), and carbon-

chlorine (C-Cl). Each of these functional groups exhibits characteristic stretching and bending frequencies, which shift upon coordination with metal ions, providing insights into the nature of the metal-ligand interactions.

The carbon-nitrogen (C=N) stretching frequency in the free ligand appears at 1601 cm^{-1} , a characteristic of imine groups. This frequency undergoes a slight shift in the metal complexes, with CuL showing a C=N stretching vibration at 1603 cm^{-1} and CuL₂ at 1606 cm^{-1} . The zinc complexes show similar shifts, with C=N stretching at 1605 cm^{-1} in ZnL and 1604 cm^{-1} in ZnL₂. These shifts reflect the coordination of the C=N group with the metal centers, reinforcing the idea that the nitrogen atoms in the imine groups are involved in metal binding. (Mahdy *et al.*, 2023). Hydroxyl (OH) stretching vibrations are present in all compounds, with the free ligand showing a peak at 3742 cm^{-1} . Upon metal coordination, the OH stretching shifts slightly, suggesting metal-hydroxyl interaction. CuL and CuL₂ show OH stretches at 3726 cm^{-1} and 3728 cm^{-1} , respectively, while ZnL and ZnL₂ exhibit OH stretching at 3725 cm^{-1} and 3722 cm^{-1} . This suggests that the hydroxyl group is likely involved in coordination with the metal ions in all complexes (Mohammad *et al.*, 2015). This peak in the complexes moves without deprotonation into lower or higher wavelength areas of $14\text{ to }20\text{ cm}^{-1}$, suggesting that oxygen is coordinated with the core metal ion. Further, Cu(II) complexes typically form in slightly acidic conditions, around pH 4-6, where the phenol is protonated, the imine coordinates, and the amine remains free. Cu(II), which favors oxygen and nitrogen donors, forms stable octahedral complexes in this pH range, with chloride and water ligands completing the coordination sphere. Zn(II) complexes, on the other hand, form in slightly less acidic to neutral conditions, around pH 5-7. In this range, Zn(II) similarly coordinates with the phenol and imine groups while chloride and water occupy the remaining sites in the octahedral geometry. Therefore, for both Cu(II) and Zn(II) complexes, the pH of formation is slightly acidic to neutral, with Cu(II) complexes forming around pH 4-6 and Zn(II) complexes forming around pH 5-7, ensuring that the phenol remains protonated and the amine uncoordinated. Carbon-oxygen (C-O) stretching vibrations are observed at 1308 cm^{-1} in the free ligand. Upon coordination with the metal ions, these frequencies shift slightly, indicating the involvement of oxygen in metal binding. CuL shows a C-O stretching vibration at 1322 cm^{-1} , while CuL₂ shifts further to 1334 cm^{-1} , suggesting stronger or different bonding in CuL₂. Similarly, zinc

complexes show C-O stretches at 1316 cm^{-1} and 1314 cm^{-1} for ZnL and ZnL₂, respectively, indicating coordination of the C-O group with the zinc ion. This change offers strong proof that the metal ion and phenolic oxygen are coordinated. The metal-oxygen (M-O) stretching vibrations confirm the formation of metal-ligand complexes. In CuL, M-O stretching is observed at 596 cm^{-1} , and in CuL₂, it shifts slightly to 591 cm^{-1} . This suggests that the oxygen atoms from the ligand are coordinating with the copper ions in both complexes, but with varying strengths or geometries. For zinc complexes, ZnL shows M-O stretching at 586 cm^{-1} , and ZnL₂ shows a shift to 588 cm^{-1} , confirming zinc-oxygen coordination in both cases. Metal-nitrogen (M-N) stretching vibrations further support the formation of metal complexes. In CuL, M-N stretching is observed at 457 cm^{-1} , while in CuL₂, it shifts slightly to 458 cm^{-1} . For the zinc complexes, M-N stretching occurs at 425 cm^{-1} in ZnL and 423 cm^{-1} in ZnL₂. These peaks confirm the involvement of nitrogen atoms in metal coordination in all the complexes, as expected from the shifts in the C=N. The new vibration band appeared in the metal complexes due to water molecules coordinated with the complexes. CuL shows a strong O-H (H₂O) stretching at 3640 cm^{-1} , indicative of coordinated water, while ZnL also shows water coordination with a peak at 3583 cm^{-1} (Al-daffay, 2023; Khalaf *et al.*, 2022).

Table 7. FTIR spectral data for imine quinoline ligand (L) and its Cu(II) and Zn(II) metal complexes

Cpds									
cm^{-1}	$\nu\text{H}_2\text{O}$	νNH_2	$\nu=\text{NH}$	$\nu\text{C-O}$	$\nu\text{C=N}$	$\nu\text{-OH}$	$\nu\text{M-O}$	$\nu\text{M-N}$	$\nu\text{C-Cl}$
L		3854	3425	1308	1601	3742			749
CuL	3640	3860	3457	1322	1603	3726	596	457	752
CuL₂		3873	3463	1334	1606	3728	591	458	739
ZnL	3583	3788	3460	1316	1605	3725	586	425	753
ZnL₂		3788	3464	1314	1604	3722	588	423	750

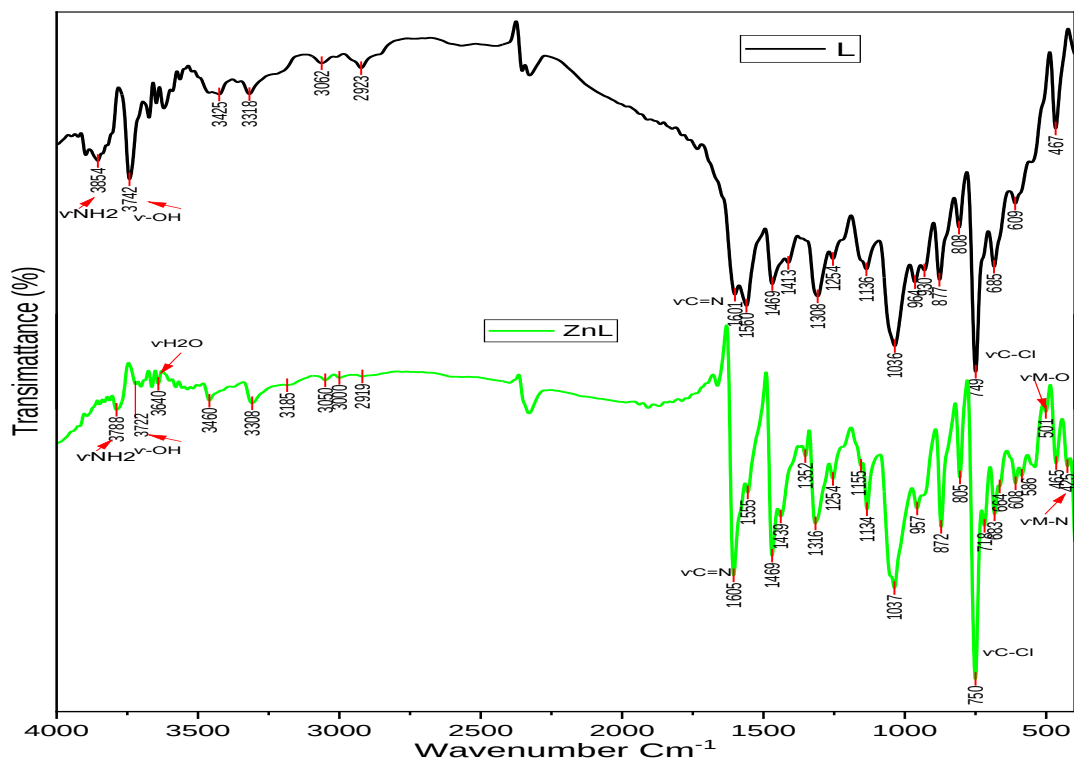


Figure 18. FT-IR spectrum of **ZnL** complexes (1:1 ratios) with the imine quinoline ligand (**L**)

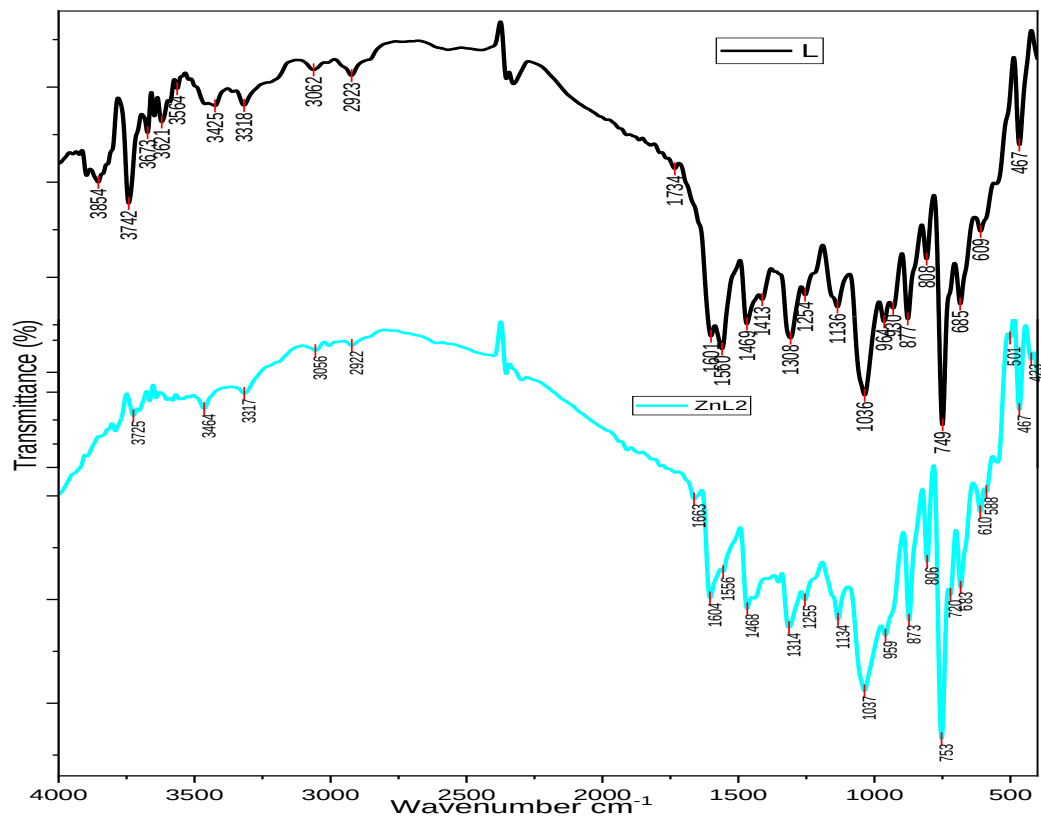


Figure 19. FT-IR spectrum of **ZnL₂** complexes (1:2 ratios) with the imine quinoline ligand (**L**)

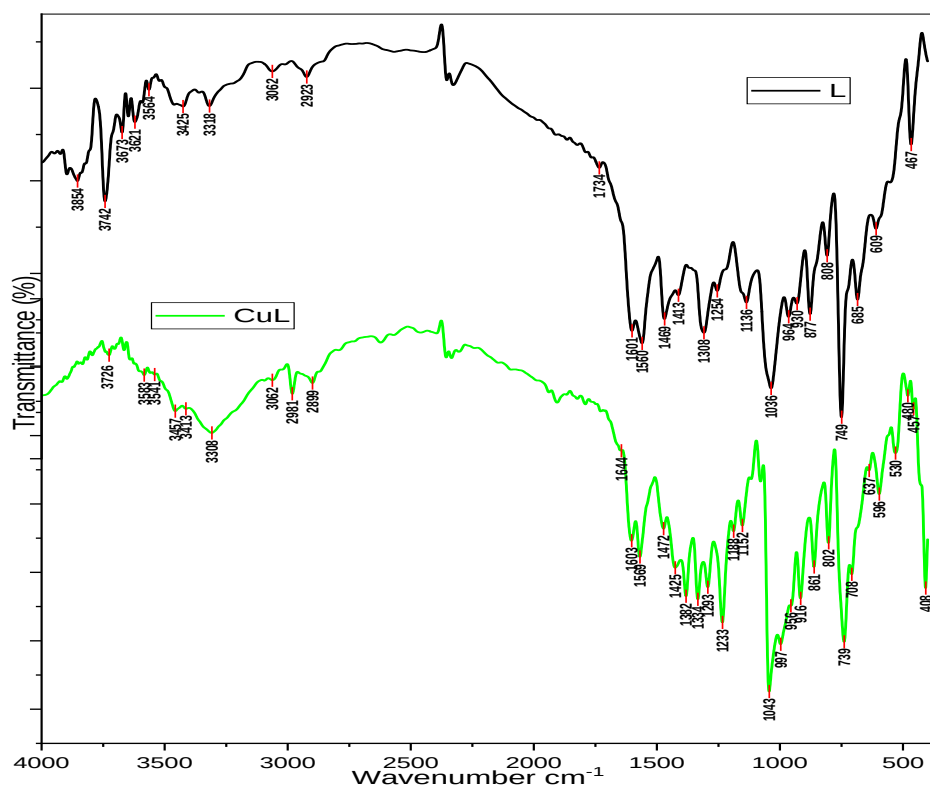


Figure 20. FTIR spectrum of Cu(II) complexes (1:1 ratios) with the ligand (L)

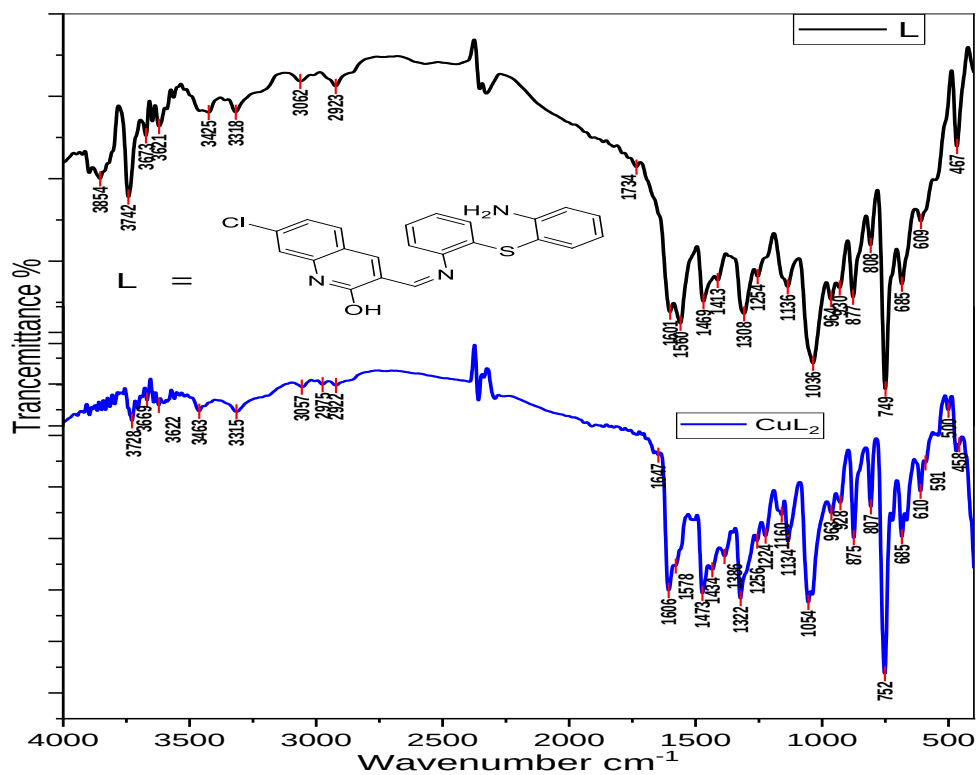


Figure 21. FTIR spectrum of Cu(II) complexes (1:2) with the ligand (L)

4.8.3. FTIR Spectra decorated metal complexes with AgNPs

The FT-IR spectra of AgNPs decorated with the imine quinoline ligand and its metal complexes includes: AgNP@L, AgNP@CuL, AgNP@CuL₂, AgNP@ZnL and AgNP@ZnL₂ were recorded in the 4000–400 cm⁻¹ range to identify the frequencies of functional groups and the vibration frequency results are shown in Table 8 and Figure 22-24. The spectral data of silver nanoparticles (AgNPs) composited with the imine quinoline ligand (L) and Cu(II) and Zn(II) metal complexes reveal interesting interactions between the AgNPs and the metal-ligand systems. While the overall spectra of the composited and uncomposited complexes remain similar, there are notable shifts in wave numbers, particularly the appearance of distinct AgNP peaks in the composited samples. These shifts provide insight into the bonding environment and coordination changes upon compositing.

The NH₂ stretching frequency remains consistent around 3794–3795 cm⁻¹ for most complexes except for AgNP@ZnL, where it is absent. This suggests that while the amino group participates in metal coordination or AgNP interaction, its bonding characteristics are largely unaffected. The C-O stretching frequency, ranging from 1299 cm⁻¹ to 1332 cm⁻¹, shows slight shifts, indicating a change in the environment around the oxygen atom when the ligand is coordinated with metal ions. The lower wave numbers, such as 1299 cm⁻¹ in AgNP@ZnL₂, suggest a stronger interaction between the metal and oxygen atoms, weakening the C-O bond slightly. The C=N stretching frequencies are relatively stable, ranging from 1601 cm⁻¹ to 1606 cm⁻¹ across the samples (Mahdy *et al.*, 2023). This implies that the imine group is actively involved in metal coordination, although the interaction with AgNPs does not significantly alter the C=N bond strength. The small shifts reflect electron delocalization and involvement of the imine group in complexation. Similarly, the O-H stretching frequencies remain around 3722–3730 cm⁻¹, indicating that the phenol group is not deprotonated and is likely involved in hydrogen bonding. The minor shifts suggest changes in the hydrogen-bonding environment due to the presence of AgNPs and metal complexes. The M-O stretching frequencies, observed in the range of 581–596 cm⁻¹ for both Cu(II) and Zn(II) complexes, confirm the formation of metal-oxygen bonds (Al-daffay, 2023; Khalaf *et al.*, 2022). The differences in

wave numbers suggest variations in metal-ligand bond strength, with Zn(II) showing slightly weaker M-O bonds compared to Cu(II). The M-N stretching frequencies fall between 460–472 cm^{-1} , indicating strong coordination between the metal and nitrogen atoms in the ligand. The shifts upon compositing with AgNPs suggest that the metals are strongly coordinated to the nitrogen atoms, especially the imine nitrogen. One of the most significant observations is the appearance of AgNP peaks, ranging from 404–443 cm^{-1} , which confirm the successful compositing of silver nanoparticles with the ligand and metal complexes. These peaks are absent in the un-composited samples. The variation in AgNP peak positions suggests different levels of interaction between the AgNPs and the metal complexes, with Cu(II) complexes showing slightly lower frequencies (441–443 cm^{-1}) than Zn(II) complexes (416–417 cm^{-1}). This implies that Cu(II) complexes may form stronger interactions with AgNPs compared to Zn(II) complexes (Shahzadi *et al.*, 2022).

Table 8. FTIR spectral data for AgNP decorated imine quinoline ligand (L) and its Cu(II) and Zn(II) metal complexes

Cpds cm^{-1}	νNH_2	$\nu\text{C-O}$	$\nu\text{C=N}$	$\nu\text{-OH}$	$\nu\text{M-O}$	$\nu\text{M-N}$	νAgNP
AgNP@L	3794	1332	1604	3728			443
AgNP@CuL	3794		1606	3728	596	472	441
AgNP@CuL₂	3793	1318	1603	3722	591	463	404
AgNP@ZnL		1323	1606	3725	592	460	417
AgNP@ZnL₂	3795	1299	1601	3730	581	469	416

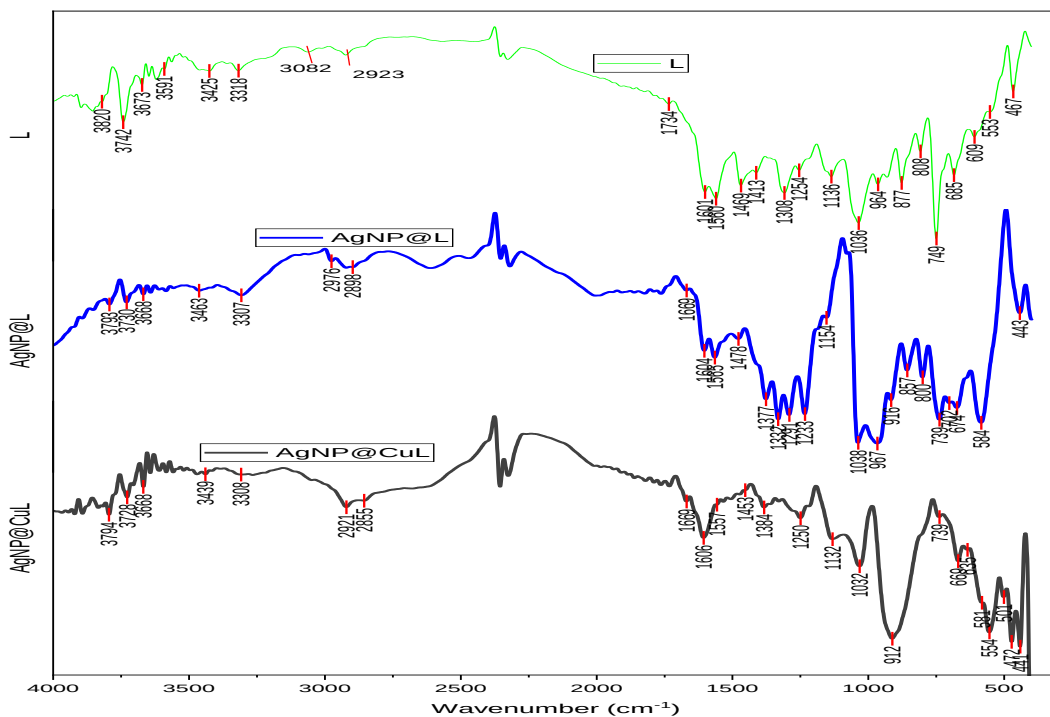


Figure 22: FTIR spectrum of synthesized L, AgNP@L and AgNP@CuL

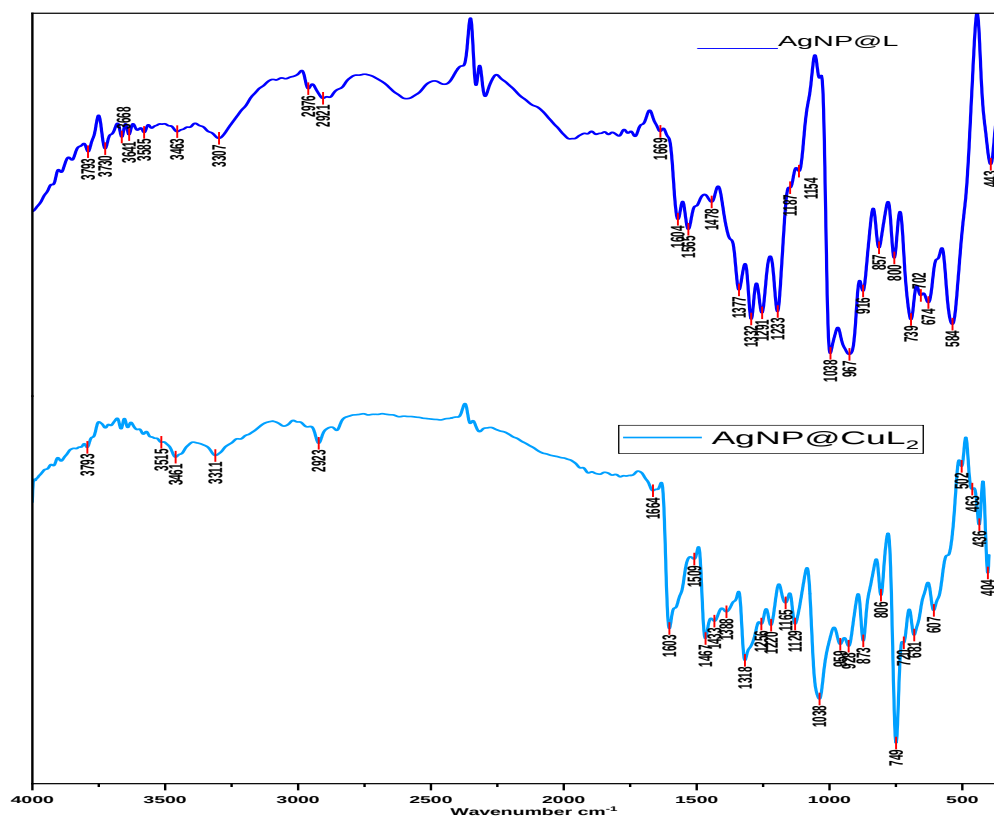


Figure 23. FTIR spectrum of synthesized AgNP@L and AgNP@CuL₂

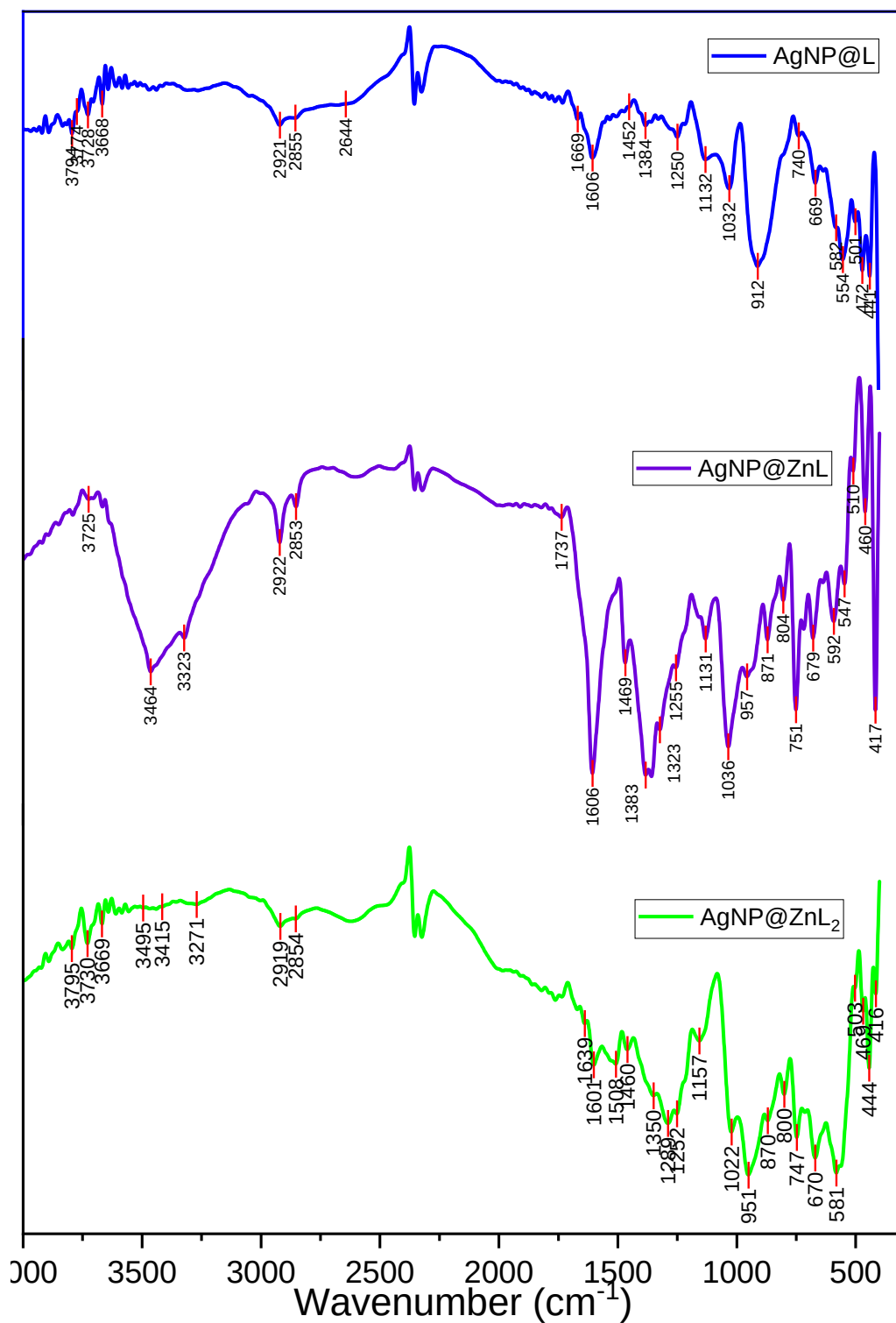


Figure 24. FTIR spectrum of synthesized AgNP@L, AgNP@ZnL and AgNP@ZnL₂

4.9. Scanning Electron Microscopy and energy dispersive X-ray analysis (SEM-EDX)

4.9.1. SEM-EDX analysis of silver nanoparticles (AgNP-His)

The biogenic AgNP-his was examined using scanning electron microscopy (SEM) that the SEM was used to analyze the compounds' morphology and particle size. The SEM micrographs showed (Figure 25C) that spherical AgNPs were heterogeneously dispersed on the glass.

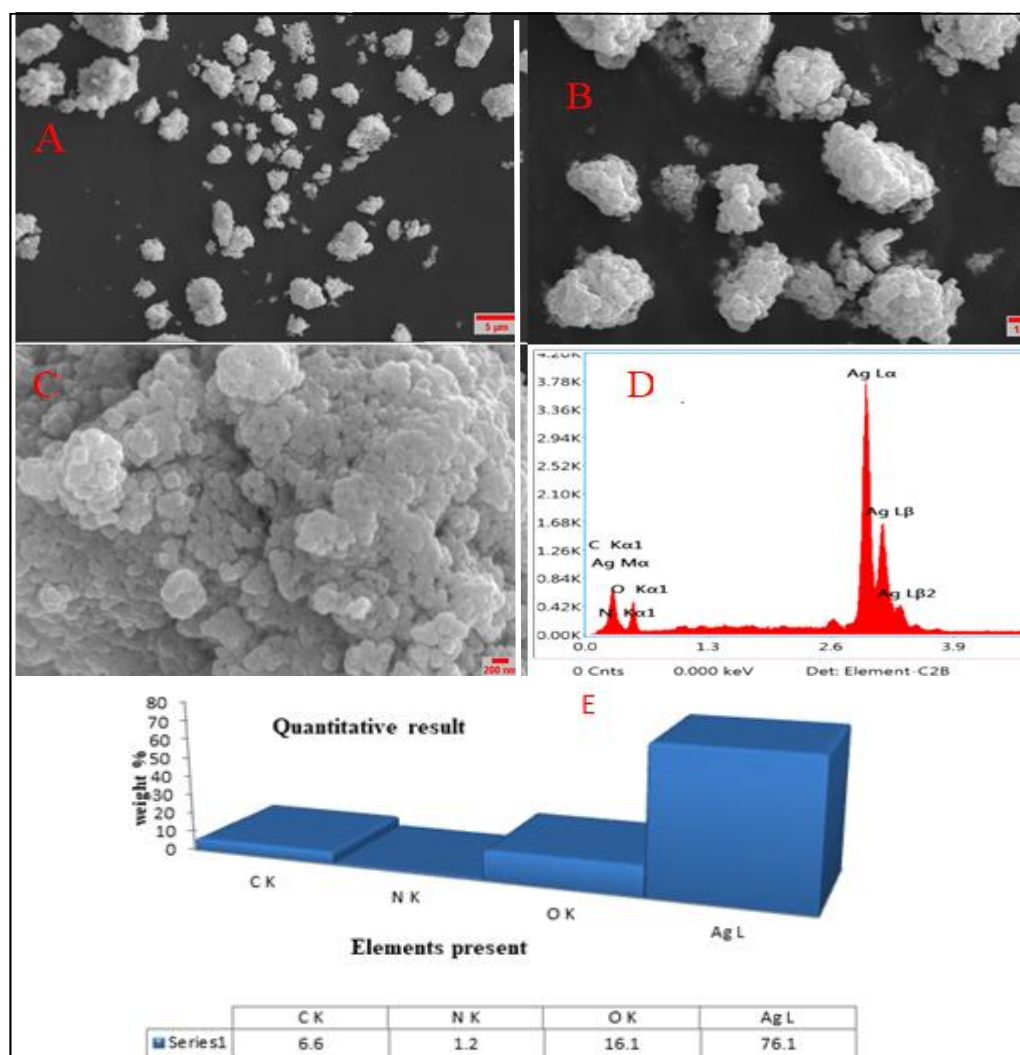


Figure 25. SEM micrograph images of synthesized AgNP-H at different magnification scale (A) 5 μm, (B) 2 μm, (C) 200 nm and (D and E) EDAX graph and elemental composition

Heterogeneous regions of high intensity are cleared in the image. No apparent quenching of these bright regions was observed, as shown in the images. These observation asserts that the heterogeneous regions can be attributed to microcrystallization of AgNPs in the supported, since microcrystals are highly emissive (Trang *et al.*, 2024). The composition of the AgNPs was obtained from EDX analysis in Figure 25 (D and E). In the EDX spectrum, AgNPs showed four signals, which correspond to C, O, N and Ag, with the amount of Ag being the highest (79.1%). The synthesized silver nanoparticles exhibited strong absorption in the 2.5 to 4 keV range. Metallic silver nanoparticles showed a typically strong signal peak at about 3 keV due to surface Plasmon (Alabdallah and Hasan, 2021). These results are in agreement with previously reported results using *Petalium nurex* (Alabdallah and Hasan 2021), *aloe Vera* and *curcumin sativus* leaf extracts (Riaz *et al.*, 2022). This proves that AgNP-his existed without any impurity. It also clearly confirmed the formation of the functionalized AgNP-His.

4.9.2. SEM-EDX analysis of the decorated silver nanoparticle with copper complexes

The biogenic AgNP-His decorated with imine quinoline ligand (L) (AgNP@L) and its Cu(II) complexes (AgNP@CuL and AgNP@CuL₂) were examined using scanning electron microscopy (SEM) that the SEM was used to analyze the compounds morphology and particle size. The SEM micrographs showed in Figure 26 (A & B) that the ligand and the AgNPs showed distinct morphology. The ligand evidenced needle like structure aggregated in a certain fashion to exhibit a flower like structure upon which the deposition of spherical aggregates of AgNP@L is observed. Heterogeneous regions of high intensity are cleared in the image. The composition of the AgNP@L was obtained from EDX analysis in Figure 26 (C and D). In the EDX spectrum, AgNP@L showed six signals, which correspond to C, Cl, O, N, S and Ag, with the amount of C being the highest (51.17%) and Ag (27.12). The decorated AgNP@L exhibited strong absorption in the 2.5 to 4 keV range corresponding to Ag. Ag showed a typically strong signal peak at about 3 keV due to surface Plasmon (Alabdallah and Hasan, 2021). The EDX analysis indicates that AgNP@L existed without any impurity with appropriate compositions. The SEM micrographs in Figure 27 (A & B) showed plate like rod structure for the complex CuL (AgNP@CuL) but this morphology was completely vanished upon composite formation with AgNPs. The aggregates of the composite with no distinct

morphology is shown in Figure 27B.. The composition of the AgNP@CuL was obtained from EDX analysis in Figure 27 (C and D). In the EDX spectrum, AgNP@CuL showed seven signals, which correspond to C, Cl, O, N, S, Ag and Cu. There strong absorption of the Cu and Ag were recorded due to orbital/shell L and M energies ref. As shown figure 27 (C and D), strong absorption about 8.04 keV was exhibited due to the presence of Cu metal with the percentage composition of (9.71%). The SEM micrographs showed in Figure 28 (A & B) that major spherical shape of AgNP@CuL₂. Similarly, the composition of the AgNP@CuL₂ was obtained from EDX analysis in Figure 28 (C and D). In the EDX spectrum, AgNP@CuL₂ showed seven signals, which correspond to C, Cl, O, N, S, Ag and Cu. The strong absorption of the Cu and Ag were recorded due to orbital/shell L and M energies as shown Figure 28 (C and D) that strong absorption about 8.04 keV was exhibited due to the presence of Cu (5.7%) metal (Ahmed *et al.*, 2018)

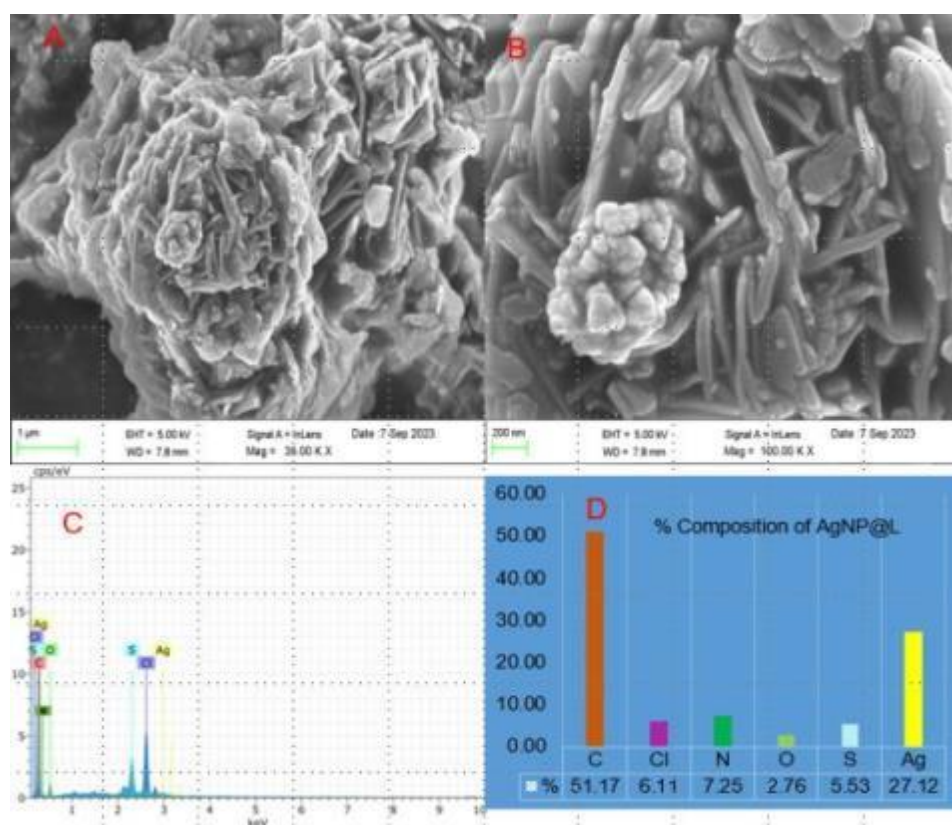


Figure 26. SEM micrograph of synthesized AgNP@L at different magnification scale (A and B) and EDX spectra (C) and Elemental composition percentage (D)

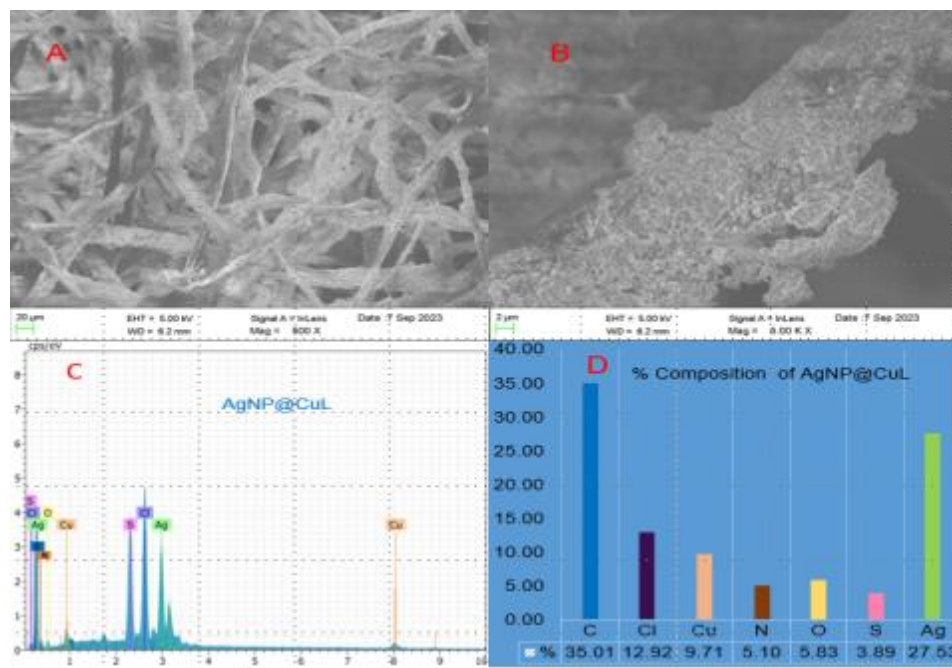


Figure 27. SEM micrograph images of synthesized **AgNP@CuL** at different magnification scale (A and B) and EDX spectra (C) and Elemental composition percentage (D)

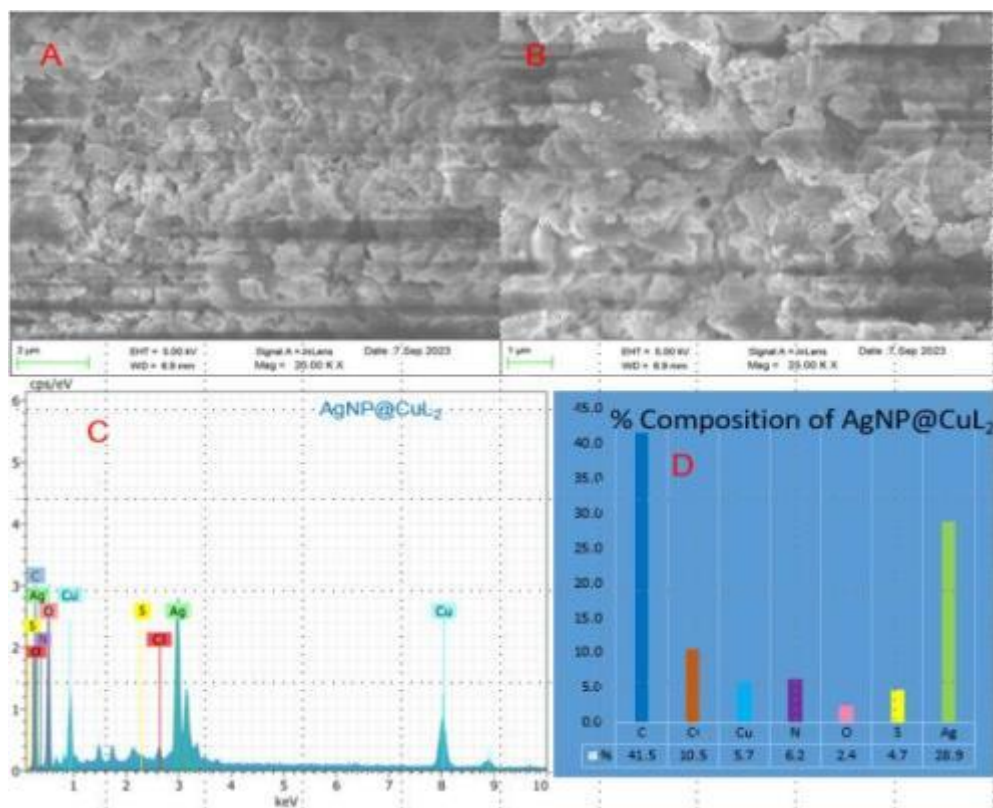


Figure 28. SEM micrograph images of synthesized **AgNP@CuL₂** at different magnification scale (A and B) and EDX spectra (C) and Elemental composition percentage (D)

The observed transition in dimensionality from 1D in AgNP@L and AgNP@CuL to 0D in AgNP@CuL₂ can be attributed to differences in the coordination and interaction of the ligands with the silver nanoparticles. In AgNP@L, this uses an imine quinolone ligand, and AgNP@CuL, with its copper complex, the 1D structures result from the ligands or complexes stabilizing or inducing needle or rod-like crystal shape of nanoparticle arrangements. The simpler coordination environment of these ligands facilitates extended structures. Conversely, the shift to 0D in AgNP@CuL₂ is likely due to the more complex coordination or multiple binding sites of CuL₂, which promotes nanoparticle clustering or aggregation into spherical or cluster-like formations. Steric effects also play a crucial role, as CuL₂ increased steric hindrance leads to a more compact, isotropic arrangement, while CuL less hindered environment supports 1D structures. Overall, the dimensional change is driven by differences in ligand coordination, steric effects, and the resulting self-assembly behaviours (Hawila *et al.*, 2023; Sharma *et al.*, 2022).

4.10. Transmission electron microscopy and HRTEM analysis of AgNPs

Transmission electron microscopy (TEM) was employed to characterize the shape, and size of the biogenic silver nanoparticle functionalized using histidine (AgNP-His). The TEM images of AgNP-his synthesized by LALE are shown in Figure 29 (A - D). The TEM image of AgNP-His indicated that the shape and the size are spherical and nanoscale respectively. AgNP-His sizes ranged from 5 to 14 nm. The difference in particle size revealed by TEM may be due to aggregation during sample preparation. The crystalline nature of AgNP-His was confirmed through the SAED pattern, which consisted of bright circular spots (Figure 29D). Spots inside the circles correspond to the inter-planar distances related to the (111), (200), (220), and (311) planes of the face-centered cubic crystalline structure of the silver nanoparticles, which were confirmed with pXRD results in this study, and related results previously reported (Alahmad *et al.*, 2022; Some *et al.*, 2019; Venugopal *et al.*, 2017). In addition, the lattice space was computed using HR-TEM, and the d-space value was about 0.382 nm as shown in Figure 30.

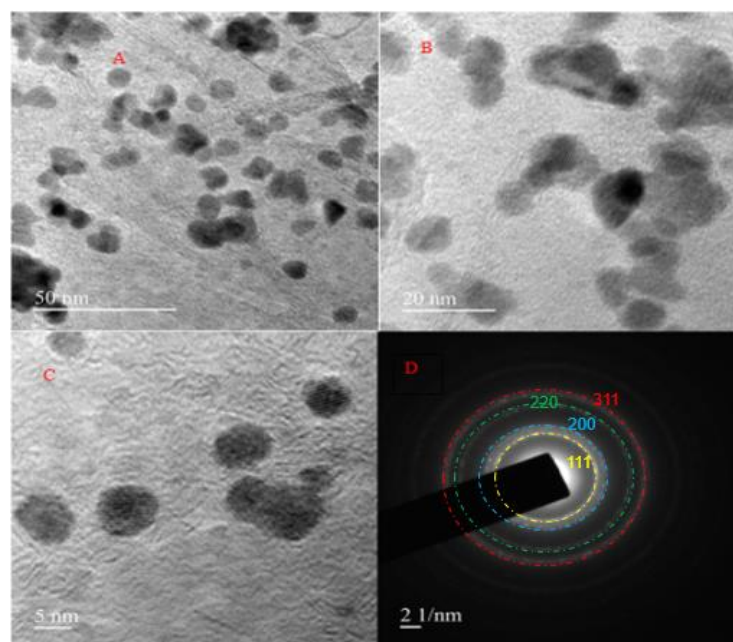


Figure 29. TEM micrograph images of synthesized AgNP-His at different magnification scales (A) 50 nm, (B) 20 nm, (C) 5 nm, and (D) SAED pattern

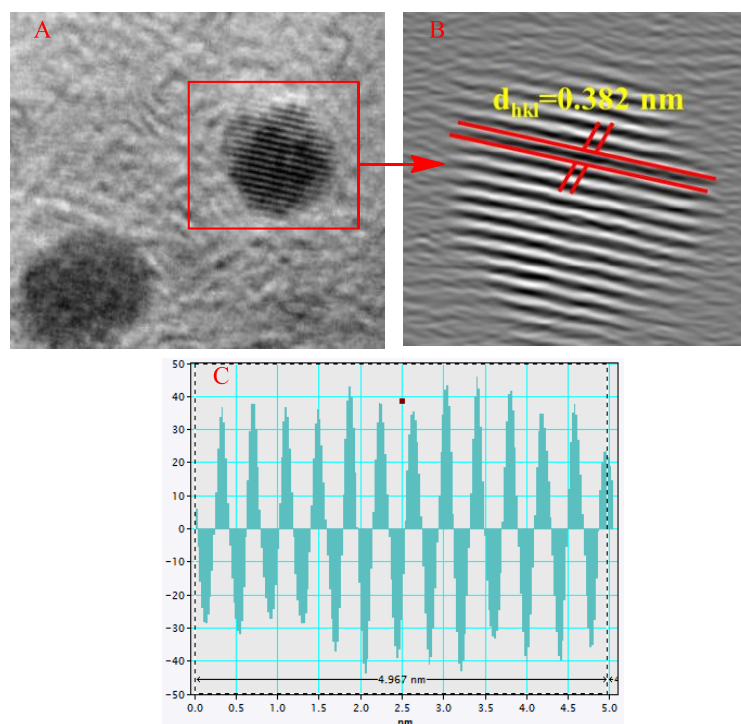
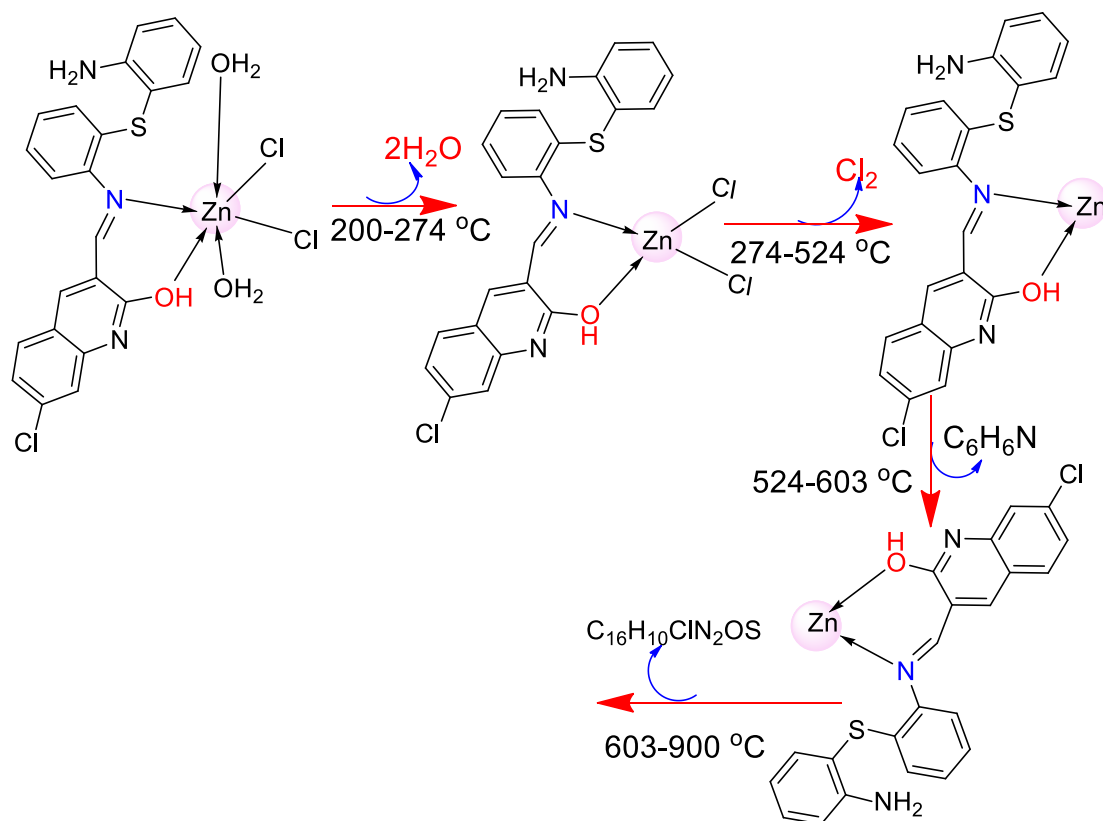


Figure 30. HR-TEM micrograph images of synthesized AgNP-His (grain boundary (A), lattice spacing (B), number of peak selected(C))

4.11. Thermal analysis

4.11.1. Thermogravimetric investigation of the zinc (II) complexes

In the present work, thermal analysis was carried out to get significant information about the thermal stability of the synthesized Zn(II) complexes and to inspect the nature of solvent molecule (if existing) to be inside the inner coordination sphere of the metal or outer sphere of it as shown in Figure 31 and Table 9. The thermogram of the ZnL complexes showed four distinct mass losses. The first one was evidenced in the temperature range from 200 -274 °C attributed to the loss of two mole of water (9.2%). first steps at a temperature range between 200 °C and 274 °C represented the loss of two mole of water (9.2%). The second step at a temperature range between 274 and 524 °C corresponding to the loss of two moles chlorine atom (14.23%). The third step at a temperature range between 524 and 603 °C the loss of C₆H₆N molecules(17.63%). The forth step at a temperature range between 603 and 900 °C the loss of C₁₆H₁₀ClN₂OS molecules (55.25%) as shown in Scheme 2.



Scheme 2. Thermal degradation mechanism of ZnL complexes

The thermogram of the ZnL_2 complexes present from decomposition three steps observed up to 900 °C: the first steps at a temperature range between 212 °C and 270 °C represented the loss of two mole of chlorine atoms (8.34%). The second step at a temperature range between 270 and 495 °C the loss of $\text{C}_6\text{H}_6\text{N}$ molecules (9.94%). The third step at a temperature range between 495 and 890 °C the loss of $\text{C}_{38}\text{H}_{26}\text{ClN}_5\text{OS}_2$ molecules (75.1%).

Table 9. The Zn(II) complexes' thermal breakdown with their suggested molecules

Complexes	Temperature range (°C)	% of removed	Proposed degraded molecules
$[\text{ZnL}(\text{H}_2\text{O})\text{Cl}_2]$	200-274	9.2	Two water molecules
$\text{C}_{22}\text{H}_{20}\text{Cl}_3\text{N}_3\text{O}_3\text{SZn}$	274-524	14.23	Two chlorine ions
MW: 578.22	524-603	17.63	$\text{C}_6\text{H}_6\text{N}$
	603-900	55.25	$\text{C}_{16}\text{H}_{10}\text{ClN}_2\text{OS}$
$[\text{ZnL}_2\text{Cl}_2]$	212-270	8.34	Two chlorine ions
$\text{C}_{44}\text{H}_{32}\text{Cl}_4\text{N}_6\text{O}_2\text{S}_2\text{Zn}$	270-495	9.94	$\text{C}_6\text{H}_6\text{N}$
MW: 948.09 g/mol	495-890	75.1	$\text{C}_{38}\text{H}_{26}\text{ClN}_5\text{OS}_2$

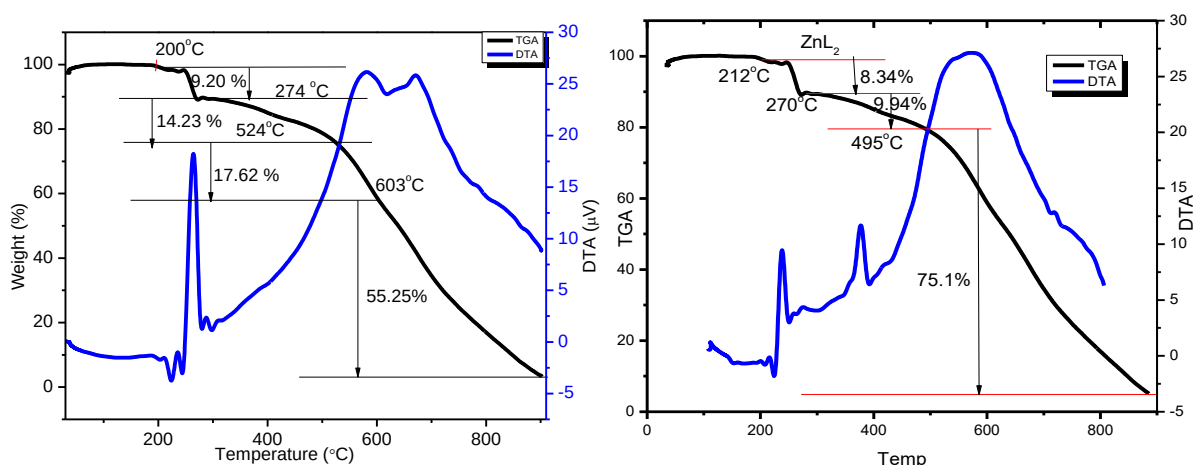
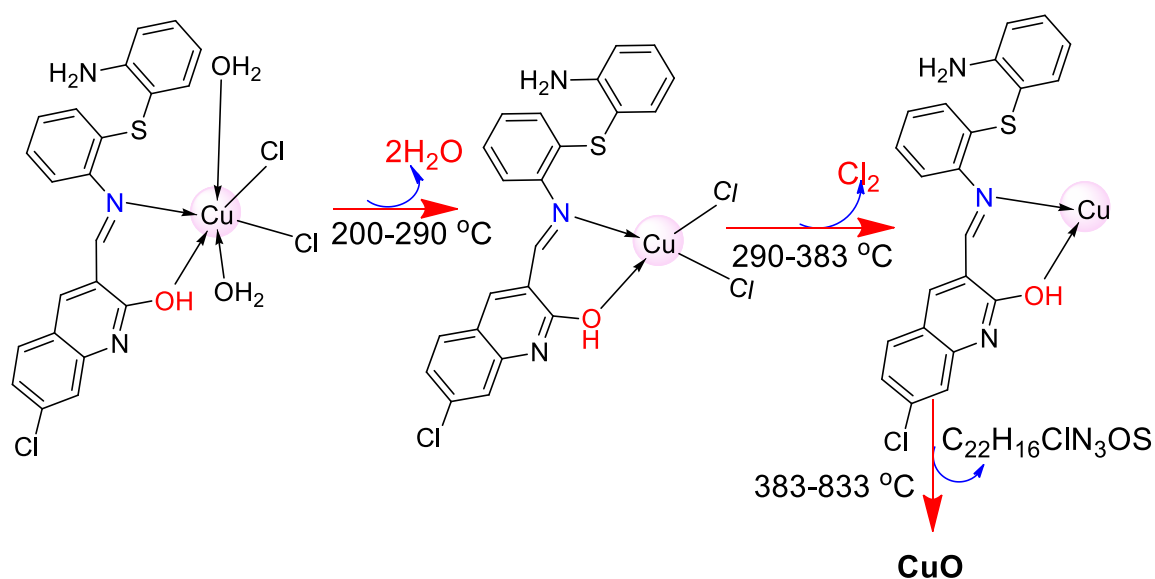


Figure 31. The TGA and DTA of Zinc Complexes a) ZnL and b) ZnL_2

4.11.2. Thermogravimetric investigation of the copper (II) complexes

In this study, thermal analysis was carried out to get significant information about the thermal stability of the synthesized Cu(II) complexes and to inspect the nature of solvent molecule (if

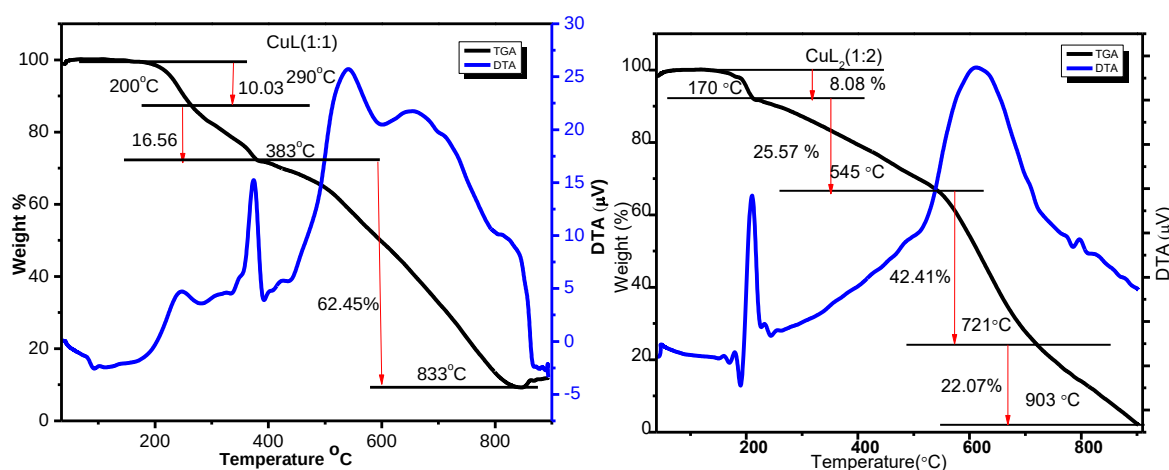
existing) to be inside the inner coordination sphere of the metal or outer sphere of it as shown in Figure 32 and Table 10. The thermogram of the CuL complexes present from decomposition three steps observed up to 833 °C that the first steps at a temperature range between 200 °C and 290 °C represented the loss of two mole of water (10.03%). The second step at a temperature range between 290 and 383 °C corresponding to the loss of two moles of chlorine atom (16.56%). The third step at a temperature range between 383 and 833 °C the loss of C₂₂H₁₆ClN₃OS molecules (10.96%) and the constant loss of mass observed at temperature > 833 °C indicated the formation of CuO as shown in Scheme 3. The thermogram of the CuL₂ complexes present from decomposition four steps observed up to 903 °C that the first steps at a temperature range between 170 °C and 225 °C represented the loss of two mole of chlorine atom (8.08%). The second step at a temperature range between 225 and 545 °C the loss of C₁₂H₁₆N₂S molecules (25.57 %). The third step at a temperature range between 545 and 721 °C the loss of C₂₂H₁₆ClN₃S molecules (42.41%). The fourth step at a temperature range between 721 and 903 °C the loss of C₁₁H₇ClNO molecules (22.07%).



Scheme 3. Thermal degradation mechanism of CuL complexes

Table 10. The Cu(II) complexes' thermal degradation with their suggested molecules

Complexes	Temperature range (°C)	% of removed	Proposed degraded molecules
[CuL(H ₂ O) ₂ Cl ₂]	200-290	10.03	Two water molecules
C ₂₂ H ₂₀ Cl ₃ CuN ₃ O ₃ S	290-383	16.56	Two chlorine ions
MW: 576.38 g/mol	383-833	62.45	C ₂₂ H ₁₆ ClN ₃ OS the ligand
	>833	10.96	CuO
[CuL ₂ Cl ₂]	170-225	8.08	Two chlorine ions
C ₄₄ H ₃₂ Cl ₄ CuN ₆ O ₂ S ₂	225-545	25.57	C ₁₂ H ₁₆ N ₂ S
MW: 946.25 g/mol	545-721	42.41	C ₂₂ H ₁₆ ClN ₃ S
	721-903	22.07	C ₁₁ H ₇ ClNO

**Figure 32.** TGA/DTA of copper complexes (CuL (1:1) and CuL₂ (1:2))

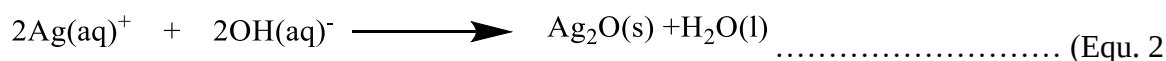
4.12. Powder X-Ray Diffraction (pXRD) Analysis

4.12.1. pXRD pattern analysis of AgNPs

The recorded diffractograms are presented in Figure 33. Three crystallographic systems for non-functionalized silver nanoparticles (AgNP), silver nanoparticles functionalized histidine (AgNP-His), and silver nanoparticles functionalized phenylalanine (AgNP-phen) were measured. The pXRD diffraction peaks of the The pXRD diffraction pattern of pristine/non-

functionalized AgNPs showed peaks at 2θ values of 32.84° , 38.18° , 44.34° , 54.92° , 64.46° , and 77.38° which correspond to the planes' *hkl* according to the Miller nomenclature: (110) (111), (200), (220), and (311) respectively. The pXRD data closely matched JCPDS No. 00-041-1104 and JCPDS Card No. 04-0783, which describe the cubic structural system of Ag/Ag₂ONPs. (Dharmaraj *et al.*, 2021; Polani *et al.*, 2017). On the other hand, both the histidine and phenylalanine functionalized AgNPs showed peaks at 2θ values 38.18° ; 44.34° ; 64.46° , and 77.5° which is equivalent to the following planes' *hkl*: (111), (200), (220), and (311) plane and 38.16° ; 44.34° , 64.48° and 77.44° which corresponds to the following planes' *hkl*: (111), (200), (220), and (311) plane respectively. These closely resemble JCPDS card No. 04-0783, which defines the cubic structural system of AgNPs (Meva *et al.*, 2016). The presence of functionalizing agents played a role in masking the possible oxidation of the as-synthesized Ag NPs.

According to the pXRD diffractogram analysis, AgNPs were found to be crystalline and contain silver ions. Besides, the peak near 32.85° (110) implied the possible existence of Ag₂O (Dharmaraj *et al.*, 2021; Fayyadh and Alzubaidy, 2021) and the peak about 54.92° corresponding to the miller index of (211). Ag ions (salt) were converted to silver (I) oxide, whereas by significant raising of the pH of the medium during the biogenic of silver nanoparticles, Ag⁺ can be converted to silver (I) oxide due to NaOH (aq) used for the pH adjustment. The use of NaOH to produce a solid Ag₂O phase was previously reported (Kayed and Mansour, 2022; Laouini *et al.*, 2021) that silver ions in the presence of OH ions form AgOH (aq), which quickly decompose when heated and precipitated to form the desired Ag₂O (Polani *et al.*, 2017) as shown in the following chemical reaction (Eqn. 2). Conversely, the use of amino acids (phenylalanine and histidine) as reducing and capping agents has significantly supported the Ag⁺ to Ag⁰ reduction (Figure 33(A and B)).



The average crystal size of the nanoparticles was computed from selected major peaks using the Debye–Scherrer's formula ($D = (k\lambda/\beta \cos \theta)$), where D represents the crystalline size (nm), λ indicates the wavelength of X-rays (0.1541 nm), β specifies the angular line full width at half maximum (FWHM) of the peak (in radians), and θ displays the Braggs angle in degrees (Giri *et al.*, 2022). The crystalline size of the biogenic AgNPs was found to be 26.64 nm,

25.63 nm, and 34.29 nm for green synthesis of AgNP, AgNP-His, and AgNP-phen, respectively (Table 11).

Furthermore, when amino acid, histidine, is added at high pH, it prevents the formation of Ag₂O and promotes the exclusive formation of AgNPs due to its chelating, reducing, and stabilizing properties. The imidazole and amine groups in histidine chelate Ag⁺ ions, making them less available to react with OH⁻ to form Ag₂O and more likely to be reduced to Ag⁰. Histidine also acts as a stabilizing agent, capping the surface of AgNPs as they form, which prevents aggregation and further reactions with OH⁻ ions. Additionally, histidine can buffer the local pH, moderating excessive alkalinity and promoting the selective reduction of Ag⁺ to Ag⁰ without intermediate oxide formation. This selective reduction mechanism ensures that AgNPs form efficiently while the strong adsorption of histidine onto the nanoparticle surface provides surface passivation, further preventing Ag₂O formation. As a result, histidine directs silver ions towards nanoparticle formation while inhibiting the formation of silver oxide, even in a highly basic environment (Kumar *et al.*, 2018).

Table 11. The average crystal size of the AgNPs was calculated from XRD spectra.

Compounds		2 θ (degrees)	FWHM (radian)	D (nm)	Average D (nm)
AgNP	1	32.85	0.3093	26.7774	26. 64
	2	38.14	0.3052	27.5414	
	3	44.32	0.3350	25.6059	
AgNP-Hist	1	38.19	0.3160	26.6029	25.63
	2	44.34	0.4356	19.6938	
	3	64.51	0.3072	30.5796	
AgNP-Phen	1	38.19	0.2450	34.3134	34.29
	2	44.37	0.2651	32.3625	
	3	64.51	0.2596	36.1870	

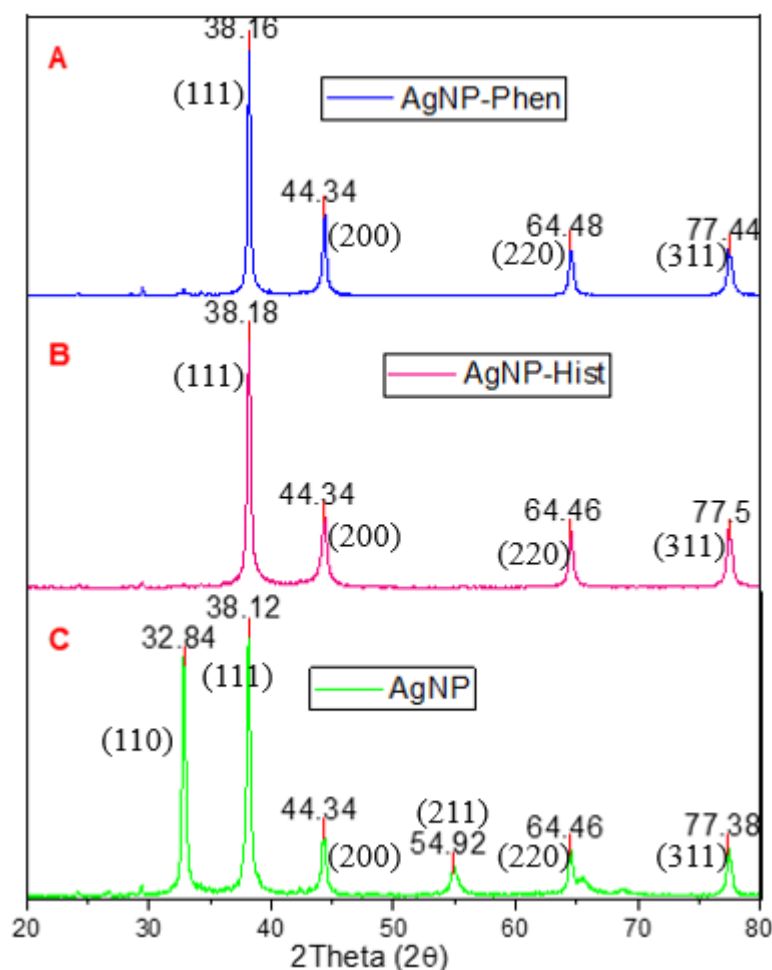


Figure 33. pXRD pattern of biogenic and phenylalanine functionalized AgNP-Phen (A), **biogenic** and histidine functionalized AgNP-Hist (B), and biogenic AgNP (C) without amino acids

4.12.2. pXRD-of the Ligand and Metal complexes

4.12.2.1. pXRD of Ligand and Zn(II) complexes

The pXRD profiles of the ligand (L), and its Zn complexes ZnL and ZnL₂ are depicted on Figure 34.. Both the ligand and the complexes exhibited polycrystalline structure. The XRD patterns of the ligand and the Zn(II) complex are more or less similar except the additional peaks emerged in the case of the complexes at 2θ values: $31.57^\circ/31.70^\circ$, $47.08^\circ/47.15^\circ$ for ZnL/ZnL₂ (Khakhlary and Baruah, 2015). The average crystal size was calculated according to Debye Scherer's formula (Equ 3):

$$d_{\text{XRD}} = 0.9\lambda/\beta (\cos\theta) \text{-----Equ (3)}$$

The equation uses the reference peaks with at angle θ where λ the wavelength of X-ray radiation and β is the width at half maximum of the reference of diffraction peak measured in degrees. The average crystalline size of L, ZnL (1:1) and ZnL₂ (1:2) were found to be 13.28, 21.19 and 26.26 nm respectively (Appendix 20). The results articulate that the complexes are nano-crystalline (Ejidike, 2018) with a potential application in biological uses.

4.12.2. pXRD of Cu(II) complexes

pXRD experiments were measured for the imine quinoline ligands and their Cu(II) complexes at room temperature are given in Figure 35. The X-ray diffraction patterns of the obtained ligand and the complexes were polycrystalline structure. Similarly, the pXRD patterns of the ligand and the CuL complex are more or less similar except the additional peaks emerged in the case of the complexes at 2θ values: 41.33° , 45.41° and 57.91° . The average crystal size was calculated using Debye Scherer's formula for the L, CuL and CuL₂. and were found to be 13.28, 31.47 and 11.54 nm, respectively (Appendix 20). The results confirmed that the complexes are nano-crystalline (Ejidike, 2018). Additionally, new XRD peaks were observed for the metal complexes at 41.33° , 45.41° and 57.91° for CuL (Khakhlary and Baruah, 2015; Nikoorazm *et al.*, 2021). However, the pXRD patterns of CuL₂ is similar to that of the ligand, which might be due to the Cu(II) metal ion masked in the ligand since it is more ratio of the ligand occurred in the sample.

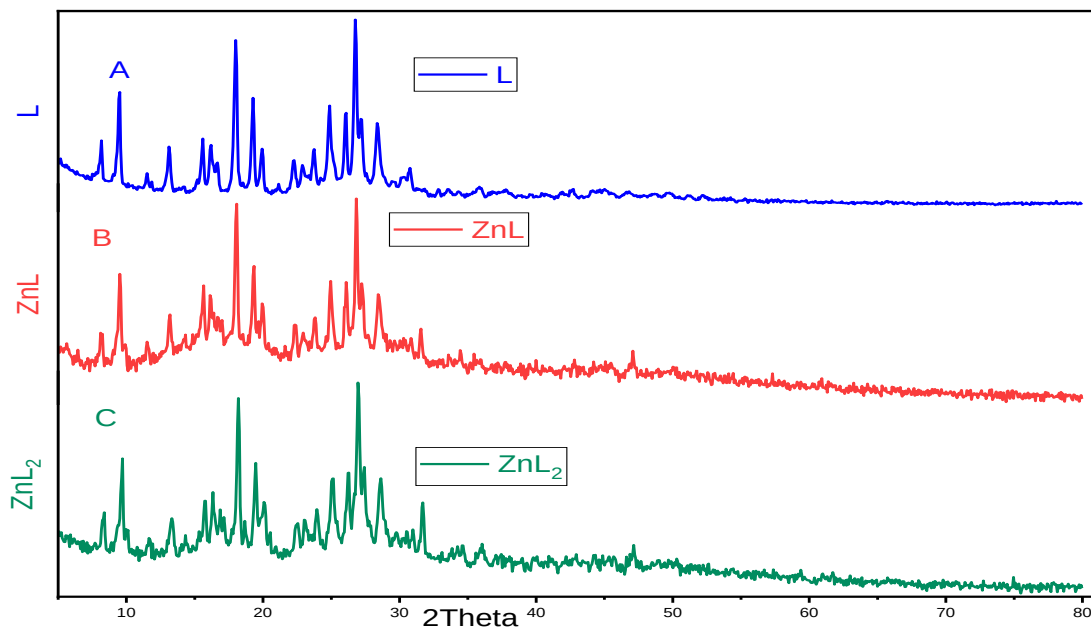


Figure 34. pXRD pattern of the ligand (L) and Zn(II) complexes

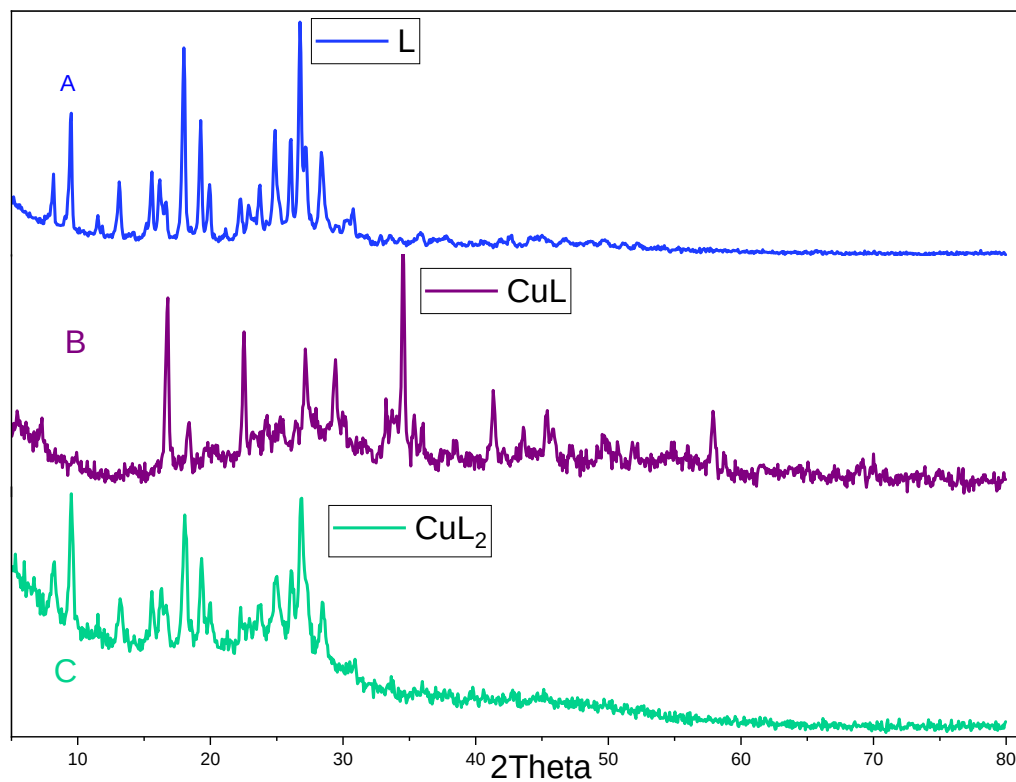


Figure 35. pXRD pattern of the ligand (L) and Cu(II) complexes

4.12.3. pXRD of the ligand and Metal complexes Decorated with silver nanoparticle

The analysis of the crystalline structure of the decorated imine quinoline ligand, CuL, CuL₂, ZnL, and ZnL₂ with silver nanoparticles (AgNPs) was performed using pXRD, as illustrated in Figures 36 and 37. The pXRD patterns for the AgNPs decorated with the ligand and its Cu(II) and Zn(II) complexes were recorded over a 2 θ range from 0° to 80°, as shown in Figure 36 (A–D). This analysis revealed five polycrystalline forms: His-AgNP@L, His-AgNP@CuL, His-AgNP@CuL₂, His-AgNP@ZnL and His-AgNP@ZnL₂. The diffraction patterns of these decorated compounds demonstrated a combination of peaks characteristic of the ligand, metal complexes, and AgNPs, as depicted in Figures 36 and 37.

The data presented in Table 12 indicates a reduction in the average crystall size for the AgNPs upon decoration with the ligand and its metal complexes. Specifically, the average crystallite sizes were found to be 26.64 nm for bare AgNPs, 20.67 nm for His-AgNP@L, 20.83 nm for His-AgNP@CuL, 19.74 nm for His-AgNP@CuL₂, 22.00 nm for His-AgNP@ZnL and 19.23 nm for His-AgNP@ZnL₂. (Appendix 20). This reduction in size can be attributed to the capping and reducing properties of the ligand and its Cu(II) and Zn(II) complexes, which play crucial roles in controlling nanoparticle growth and stabilizing dispersion. The ligand and complexes likely act as effective capping agents, promoting the formation of smaller, well-dispersed nanoparticles. These capping effects reduce agglomeration, leading to fine-tuned particle growth and enhance control over the physicochemical properties of the nanoparticles, as verified by the XRD patterns of the decorated complexes. Such control is particularly valuable in nanomaterial, where the precise tailoring of particle size and morphology is crucial for achieving specific properties and functionalities.

Table 12. The average crystal size of the decorated Ligand and its Cu(II) and Zn(II) complexes with AgNPs as calculated from XRD pattern.

Major peak AgNPs	2 θ (degrees)	FWHM (radian)	D (nm)	Average D (nm)
AgNP	32.85	0.3093	26.7774	26.64
	38.14	0.3052	27.5414	
	44.32	0.3350	25.6059	
AgNP@L	18.18	0.26	31.29	20.67
	27.02	0.62	13.22	
	29.75	0.26	31.66	
	38.47	0.43	19.72	
	44.65	0.64	13.46	
	64.78	0.50	18.91	
	77.72	0.62	16.41	
	77.72	0.62	16.41	
AgNP@CuL	26.37	4.17	1.96	20.83
	29.59	0.17	47.56	
	38.34	0.38	22.36	
	44.50	0.58	14.84	
	77.61	0.58	17.43	
AgNP@CuL ₂	27.13	2.56	3.19	19.74
	29.65	0.19	42.46	
	32.14	0.66	12.47	
	38.37	0.34	24.47	
	44.55	0.47	18.43	
	77.61	0.58	17.42	
AgNP@ZnL	16.39	0.48	17.29	22.00
	28.00	0.23	36.94	
	32.41	0.23	37.14	
	38.32	0.22	40.26	
	49.86	75.68	0.12	
AgNP@ZnL ₂	75.51	35.52	0.29	19.25
	32.23	0.65	12.64	
	38.44	0.28	29.84	
	44.62	0.34	25.14	
	46.03	1.20	7.20	
	77.68	0.48	21.42	

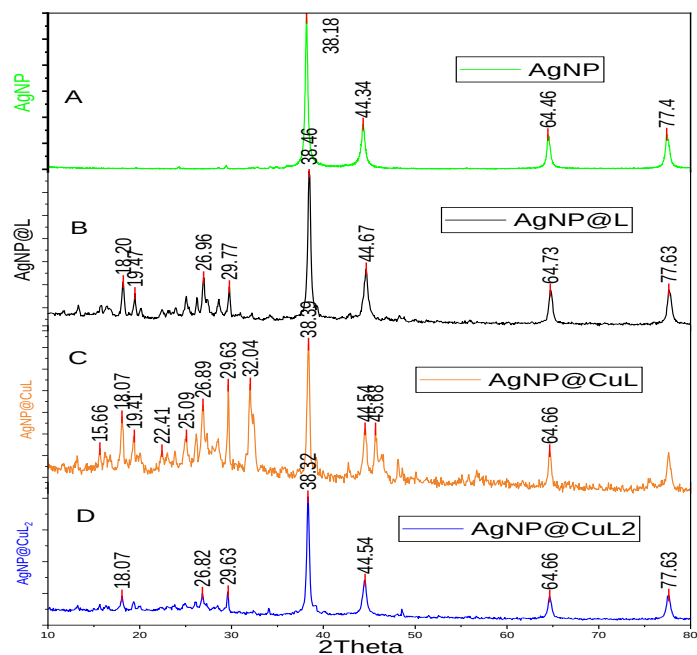


Figure 36. Powder XRD pattern of A) AgNP B) AgNP@L, C) AgNP@CuL and D) AgNP@CuL₂

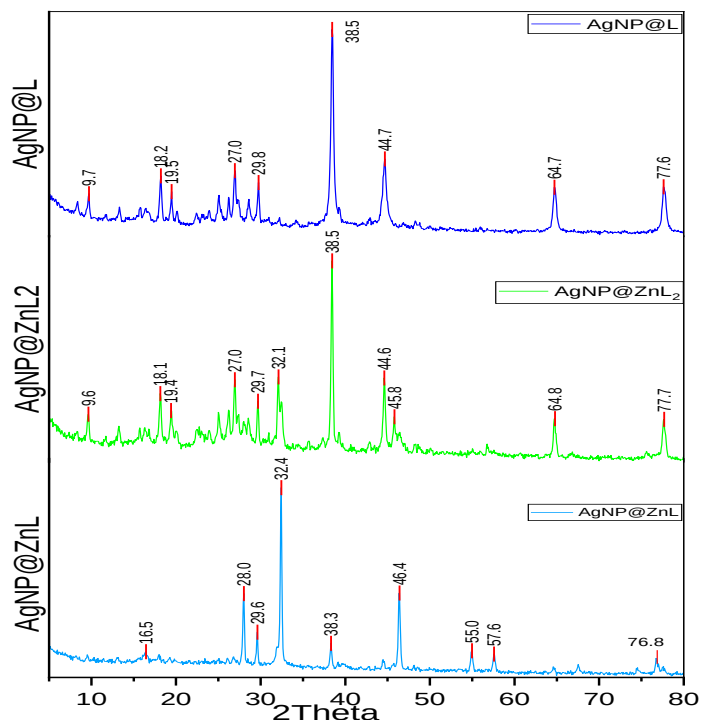


Figure 37. Powder XRD pattern of AgNP@L, AgNP@ZnL and AgNP@ZnL₂

4.13. Quantum Chemical Analysis

The results for the quantum chemical descriptors, including the band gap energy, as well as the HOMO and LUMO Eigenvalues displayed in Table 13. These descriptors have been found to exhibit significant correlations with various biological activities, such as cytotoxicity, antioxidant, and antibacterial properties. For the ligand, the band gap energy (eV) was estimated to be 3.409, whereas 3.255 for CuL₂, and 2.062 for CuL with global softness values of 0.293, 0.307, and 0.485 eV, respectively, for L, CuL₂, and CuL. The result reveals that the metal complexes exhibited a decrease in band gap energy and an increase softness.

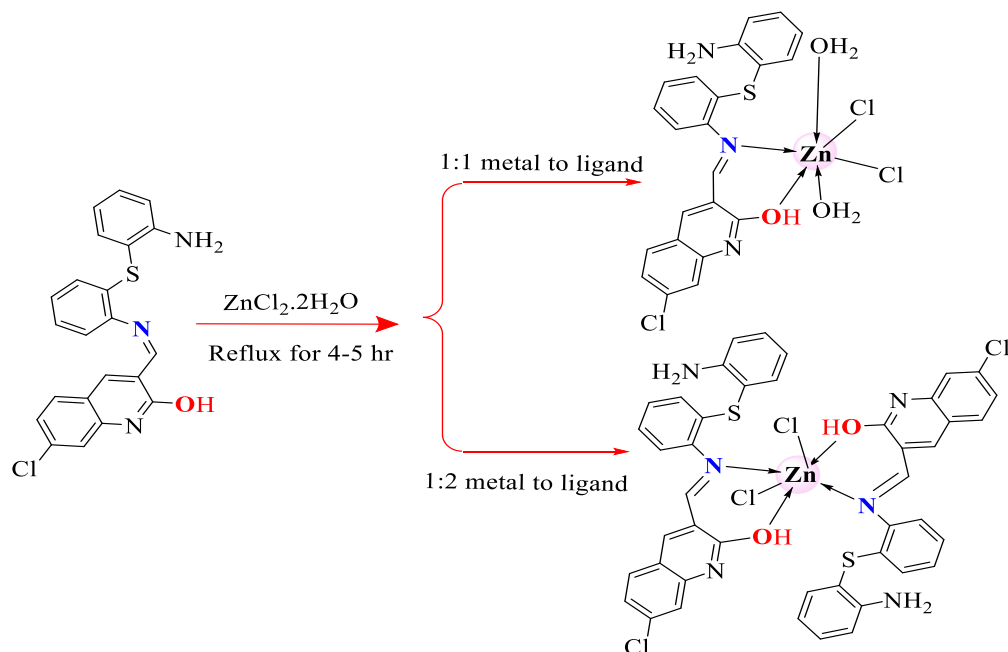
Table 13. Quantum chemical descriptors (in eV) of the ligand and its Cu(II) complexes

	HOMO	LUMO	E_g	χ	μ	η	σ	ω	Nu
IQL	-6.02438	-2.61531	3.409	4.320	-4.320	1.705	0.293	5.474	0.183
CuL	-5.87308	-3.81126	2.062	4.842	-4.842	1.031	0.485	11.372	0.088
CuL₂	-6.42165	-3.16700	3.255	4.794	-4.794	1.627	0.307	7.062	0.142

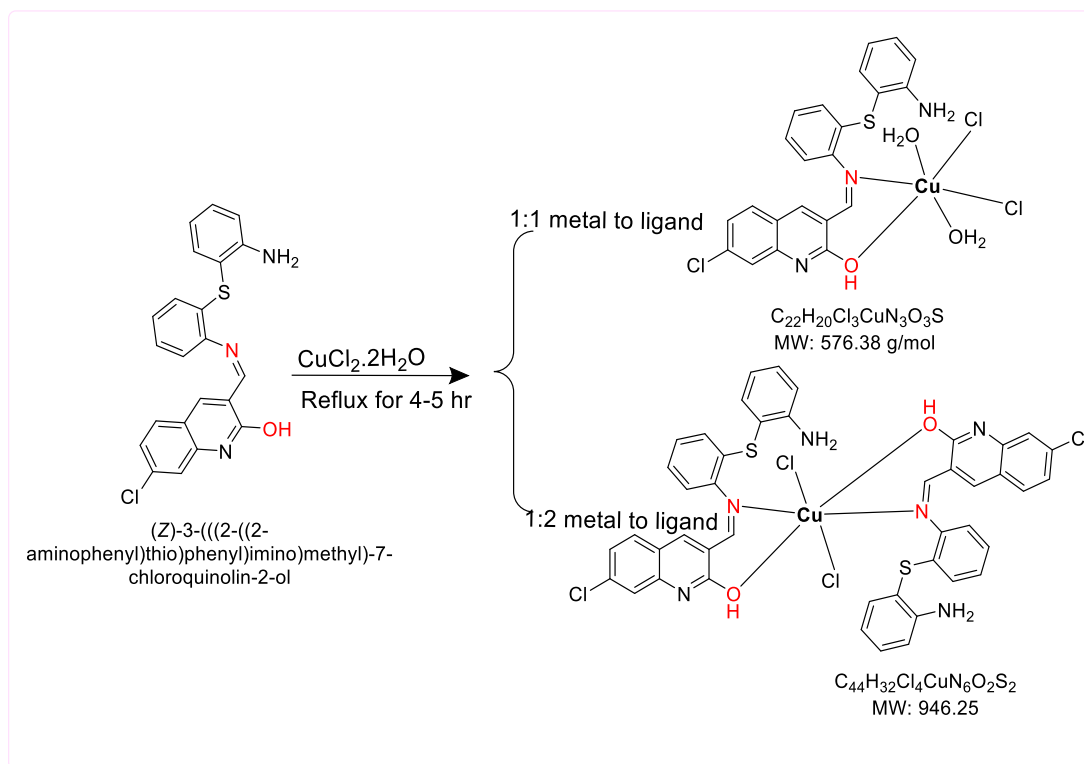
4.14. Proposed molecular structures and geometry

Generally, the resulting Zn(II) and Cu(II) complexes are solids infusible at high temperatures and stable at normal temperatures as shown in the TGA-DTA data. The metal complexes exhibit extremely low molar conductance. The metal complexes are nonelectrolytes, and do not generate any white residue with AgNO₃, as confirmed by the coordination of the chloride molecules with the metal. The bands that predominate in the diamagnetic Zn(II) complex of the imine quinoline ligand (L) are charge transfer bands. These bands may be found in an octahedral environment and range in wavelength between 377 to 460 nm. Similarly, the electronic spectra of the [ZnL₂Cl₂] complex shows an absorption band at 397 nm that is ascribed to the L→M transition, confirming the octahedral structure of the complex. Furthermore, the absorption band that supports the proposed structure is consistent with the results of the TD-DFT computation. Based on spectroscopic and analytical data, the proposed structures are given in Scheme 4 and 5. Our analysis is in agreement with previously published

results (Ahmad *et al.*, 2023; Jazzazi *et al.*, 2024; Karthika *et al.*, 2020; Loginova *et al.*, 2011; Odularu, 2023; Reddy & Reddy, 1996; Turan & Şekerci, 2010; Yernale and Mathada, 2020).



Scheme 4. The proposed molecular structures and geometry of Zn(II) complexes



Scheme 5. The proposed molecular structures and geometry of Cu(II) complexes

4.15. Biological activities Analysis

4.15.1. Anticancer activities of L and its Cu(II) complexes

The cytotoxic activities of imine quinoline ligand (L) and its Cu(II) complexes were investigated *in vitro* against the human breast cancer cell line (MCF-7) at different concentration including (6.125, 12.5, 25.0, 50.0, 100.0, 200 µg/mL) using MTT assay to determine cell growth inhibition as percent of control and computing half-maximal inhibitory concentration (IC₅₀) (50% growth inhibition). Standard anticancer drug cisplatin was used as reference compound. The results are given in Table 14 and Figure 38.

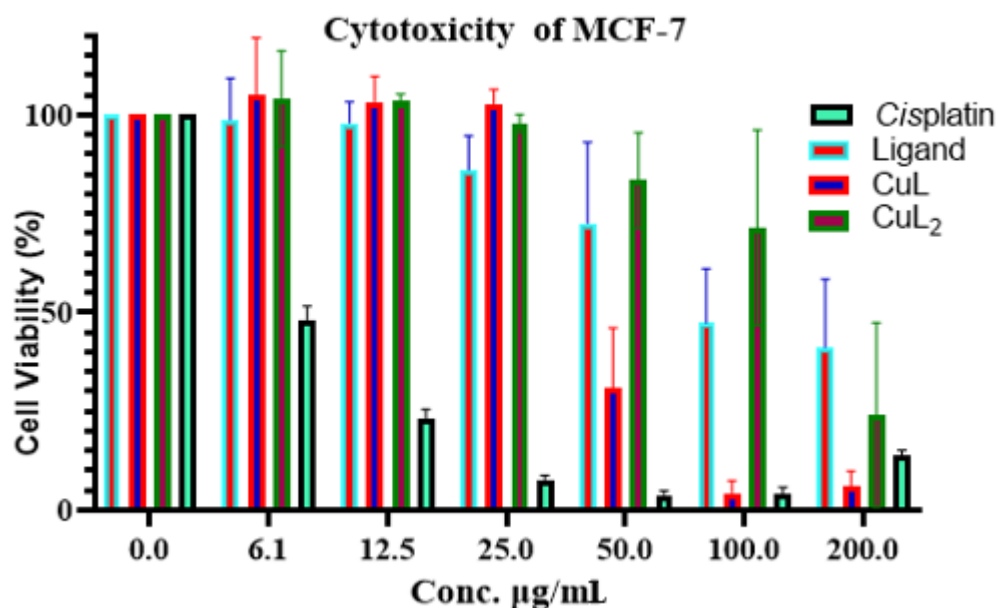


Figure 38. Dose response effect of ligand (L), cisplatin, CuL, and CuL₂ on the viability of the MCF-7 cancer cell lines

As can be seen in Figure 38 and Table 14 imine quinoline ligand (L), cisplatin, CuL and CuL₂ exhibited anti MCF-7 cell line activities *in vitro* with different degrees of potency in dose-dependent manner (Alam *et al.*, 2023; Manvatkar *et al.*, 2023). The percentage growth inhibition was found be increasing with increasing concentration of test compounds. The CuL complexes exhibited moderate cytotoxicity with IC₅₀ value $43.82 \pm 2.351 \mu\text{g/mL}$ (González-Larrazza *et al.*, 2020). Whereas CuL₂ and L exhibited weak cytotoxicity with IC₅₀ values of $132.20 \pm 5.687 \mu\text{g/mL}$ and $117.60 \pm 1.098 \mu\text{g/mL}$, respectively. The sensitivity of L,

Complexes and cisplatin in killing MCF-7 cancer cells decreases in order cisplatin > > CuL > CuL₂ > L

Table 14. Net growth as present of control and IC₅₀ value for ligand, cisplatin and Cu(II) complexes against *MCF-7* cancer cell line

$\mu\text{g/mL}$	Ligand	CuL	CuL ₂	Cisplatin
6.125	98.86 $\pm$ 8.54	94.72 $\pm$ 3.72	97.44 $\pm$ 6.16	48.28 $\pm$ 2.69
12.5	98.02 $\pm$ 4.32	97.75 $\pm$ 1.49	98.34 $\pm$ 0.59	23.37 $\pm$ 1.73
25	86.11 $\pm$ 7.04	95.55 $\pm$ 3.28	97.90 $\pm$ 1.91	7.74 $\pm$ 0.90
50	72.67 $\pm$ 16.70	31.10 $\pm$ 12.30	83.47 $\pm$ 9.82	3.85 $\pm$ 0.85
100	47.33 $\pm$ 11.28	4.37 $\pm$ 2.58	71.48 $\pm$ 20.24	4.13 $\pm$ 1.38
200	41.18 $\pm$ 14.10	6.01 $\pm$ 3.22	24.14 $\pm$ 19.06	13.78 $\pm$ 1.11
IC₅₀	117.6 $\pm$ 1.098	43.82 $\pm$ 2.351	132.2 $\pm$ 5.687	5.69 $\pm$ 1.496

Although all the synthesized compounds were moderately cytotoxic against *MCF-7* cells, their cytotoxic profiles demonstrated less potency compared to *cisplatin*, which had IC₅₀ values of 5.69 $\mu\text{g/mL}$.

4.15.2. Antibacterial activities Analysis

4.15.2.1 Antibacterial activities of biogenic AgNPs and its functionalized amino acids

In this study, two clinical strains of bacterial pathogens, *S. aureus* and *E. coli* were investigated for their *in vitro* interaction with biogenic pristine AgNPs and its histidine and phenylalanine derivatives of the same. Growth inhibition zone of the the AgNPs was compared th positive control drug of gentamycin. The solution of the nano particles was made in DMSO, which exhibited no bacterial growth inhibition and used as negative control. The results reported biological nature of biogenic AgNPs and its functionalized histidine and phenaylalanine amino acids derivatives with moderate antibacterial activity. Particularly, AgNP-H showed the maximum zone of inhibition (11.00 $\pm$ 0.82 mm) against *E. coli*, which was significantly higher as compared to the other nanoparticles (Table 15 and Appendix 12). AgNPs has shown lower activity against both bacterial strains compared to AgNP-His and

AgNP-Phen. The observation reveals that the nanoparticles are more active against *E. coli* and moderate activities against *S. aureus* bacteria. This enhanced activity may be attributed due to chelation of AgNPs with histidine and phenylalanine that provide against the bacterial strains (Al-Amiery *et al.*, 2012). The sensitivity of compounds in killing both bacterial strain decrease in the order gentamycin > AgNP-his > AgNP-phen > AgNPs.

Silver nanoparticles synthesized with extracts of *Eucalyptus camaldulensis* and *Terminalia arjuna* showed a zone of inhibition with 16 mm in diameter. The inhibition was significantly higher on gram-positive bacteria than on gram-negative bacteria (Liaqat *et al.*, 2022). However, the present results showed that AgNP functionalized with histidine was the highest and showed significantly higher activity against the gram-negative bacteria (*E. coli*). Of course, antibacterial activity and mechanism of inhibition depend on the type of the species/strain of bacteria, the nanoparticles, concentration, method of synthesis, particle size, shape and specific surface area (Wang *et al.*, 2017).

Table 15. Antimicrobial activities of the 62.5 µg/mL of 2 mM AgNO₃ of biogenic synthesized silver nanoparticles and functionalized with histidine and phenylalanine

Treatments	Antibacterial activities (Zone inhibition diameter, mm)	
	<i>S. aureus</i> ATCC 25926	<i>E. coli</i> ATCC 25922
AgNP	8.00±0.82 ^e	8.67±0.47 ^e
AgNP-His	8.67±1.25 ^e	11.00±0.82 ^d
AgNP-Phen	8.33±0.47 ^e	8.33±1.25 ^e
Gentamycin (Positive control)	22.33±0.47 ^a	20.67±0.47 ^{ab}
DMSO (Negative control)	ND	ND

Inhibition zones (mm) designated by the different letters across the rows and the columns are significantly different from each other at α=0.05 for antibacterial activities

4.15.2.2 Minimum Inhibitory Concentration (MIC) of AgNPs and its amino acid functionalized AgNPs

The MIC of the AgNP and its histidine and phenylalanine functionalized AgNPs were evaluated and the results were tabulated in (Table 16). The MIC of the AgNPs and AgNP-Phen was 25 µg/mL for *S. aureus* ATCC 25926 and *E. coli* ATCC 25922 (Table 16). AgNP-His was similar result against *S. aureus* ATCC 25926 and strong (12.5 µg/mL) against *E. coli* ATCC 25922 compared to the gram-positive bacterial pathogens tested in this study.

The minimum inhibitory concentration of AgNP-His was tested against the *E. coli* strain, which showed the highest susceptibility to biogenic AgNP-His. In this regard, the MIC of the biogenic AgNPs against *E. coli* strain was found to be 12.5 µg/mL. Interestingly, the detected MIC value of biogenic AgNPs against *E. coli* strain was lower than that detected in a prior report which demonstrated MIC value of 300 µg/mL indicating the high efficiency of synthesis method followed in our current investigation (Liu *et al.*, 2022). The biogenic silver nanoparticles utilize several mechanisms of actions, contributing to their potent antibacterial effectiveness. Here are four of AgNPs' documented antibacterial effects: (1) adherence to the microbial membrane's surface, (2) AgNPs disrupt cellular biomolecules after internalization into bacterial cells resulting in intracellular damage, (3) they cause cellular toxicity by producing reactive oxygen species (ROS) that create oxidative stress in the cell, and (4) they interfere with the cell's signal transduction pathways (Aljeldah *et al.*, 2023; Dakal *et al.*, 2016).

Table 16. The minimum inhibition concentration of the ligand and its AgNPs and its amino acid functional.

Test Samples	MIC for different Pathogenic Bacterial Strains	
	<i>Staphylococcus aureus</i> ATCC 25926	<i>Escherichia coli</i> ATCC 25922
AgNP	25	25
AgNP-His	25	12.5
AgNP-Phen	25	25
Gentamycin (Positive control)	1.688	1.688
DMSO (Negative control)	ND	ND

4.15.2.3. Antibacterial activities of imine-quinoline ligand (L) and its Cu(II) and Zn(II) complexes

The synthesized imine quinoline ligand (L) and its Cu(II) and Zn(II) complexes were evaluated for their antibacterial activity using the agar-well diffusion technique; the findings are displayed in (Table 17 and Appendix 13). The metal complexes and the synthetic ligand inhibited Gram-positive (*S. aureus*) and Gram-negative (*E. Coli* and *P. aeruginosa*) bacterial pathogens. The results showed that metal complexes had greater antibacterial activity than the ligand (L) at 50 µg/mL (Table 17). Specifically, ZnL exhibited a maximum zone of inhibition (9.50 ± 0.41 mm) against Gram-Positive (*S. aureus* ATCC 25926) than the other metal complexes. Similarly, the ZnL₂ demonstrated the highest zone of inhibition (9.67 ± 0.47 mm) against Gram-Negative (*E. Coli* ATCC 25926) and (9.33 ± 0.47 mm) against (*P. aeruginosa* ATCC 25926) than the ligand (Table 17). As shown in Table 17, the ligands did not show a significant variation of growth inhibiting between gram positive and gram-negative bacteria. Similarly, the Metal complexes did not show significant variation in antagonism, although ZnL₂ against *E. coli* recorded a relatively higher growth inhibition.

On other hand, the ligand and its Cu(II) complexes exhibited antagonistic effects against both gram-positive and gram-negative bacteria, as presented in Table 17 and Appendix 13. The inhibition zones ranged from 7.33 mm to 9.01 mm, with the Cu(II) complexes demonstrating significantly higher antagonism compared to the ligand at $\alpha = 0.01$. Notably, **CuL** displayed the highest inhibition zone (9.01 mm) against the gram-negative bacteria *P. aeruginosa*. The antibacterial activities of **CuL** were consistent across both gram-negative and gram-positive bacterial strains, ranging from 8.04 mm to 9.01 mm (Table 17). **CuL** and **CuL**₂ exhibited stronger inhibition against gram-positive bacteria (*S. aureus*) compared to gram-negative bacteria.

Overall, the antibacterial activity test revealed that the copper complexes displayed better antibacterial activity than the ligand. Specifically, **CuL** demonstrated significantly larger inhibition zones against all tested bacterial pathogens compared to **CuL**₂ (Table 17). In general, the integration of synthesized imine quinoline into the metal complex did not show

significant improvement in the antibacterial potential of the products. Similar to the current result, (Damena *et al.*, 2022b) found that 2-((2-hydroxyethyl)amino)quinoline-3-carbaldehyde (H₂L) in a 1:2 (Zn(II) and Cu(II)-to-ligand ratio) in methanol has found lower antibacterial activity against similar pathogens. This could be associated with metal complexes chelation theory that relates the ability of chelation with the passage of complexes through a cell membrane (Chebout *et al.*, 2022).

Table 17. Antimicrobial properties of the ligand and its Cu(II) and Zn(II) complexes

Molecules (50 µg/mL)	Zone inhibition diameter (mm)		
	Bacteria		
	Gram-Positive bacterial	Gram-Negative bacterial	
	<i>S. aureus</i>	<i>E. Coli</i>	<i>P. aeruginosa</i>
Ligand	7.47±0.41	7.33±0.47	7.67±0.47
CuL	8.67±0.47	8.00±0.82	9.00±0.82
CuL₂	8.33±0.47	7.33±0.47	7.67±0.47
ZnL	9.50±0.41	9.17±0.24	9.00±0.82
ZnL₂	8.83±0.24	9.67±0.47	9.33±0.47
Gentamycin (Positive control)	18.33±0.47	17.00±0.00	17.67±0.47
DMSO (Negative control)	ND	ND	ND

4.15.2.4. Minimum Inhibitory Concentration (MIC) of L and its Cu(II) and Zn(II) Complexes

The MIC of the quinoline ligand and its Cu(II) and Zn(II) complexes evaluated and the results were tabulated in (Table 18). The MIC of the Ligand and CuL was 25 µg/mL and 12.5 µg/mL, respectively, for the tested bacterial pathogens (Table 18). CuL₂ was strong against *S. aureus* ATCC 25926 compared to the gram-negative bacterial pathogens tested in this study. Similarly the literature reports for the copper complexes are stronger against gram positive bacteria as compared to gram-negative bacteria (Alem *et al.*, 2023). However, unlike a previous report, (Alem *et al.*, 2023) the present Cu(II) complexes inhibited gram positive bacterial strains at 12.5 µg/mL although the minimum inhibition zone did not coincide. In

general, the complexes' ability to limit bacterial growth was not improved by the integration of ligand and copper metal. Previous study also showed that there is a lesser inhibitory potential of the Ligands in comparison to a pure metal complex (Al-Qadsy *et al.*, 2023).

Similarly, the MIC of the metal complexes (ZnL and ZnL₂) was 12.5 µg/mL for the strains of gram-negative bacteria (*Pseudomonas aeruginosa* ATCC 248 and *Escherichia coli* ATCC 25922) (Table 18). This indicates that gram negative bacteria are more susceptible to the metal complexes' effects than gram positive bacteria (Table 18). Similar results were reported by (Chkirate *et al.*, 2020) where zinc complexes inhibit the two Gram negative bacterial species at 12.5 µg/mL. However, the integration of ligands with zinc metal didn't show improvement in the bacterial growth inhibition potential of the complexes. The relative lower inhibition potential of other ligands as compared to the pure metal complex is also reported by (Al-Qadsy *et al.*, 2023).

Table 18. The minimum inhibition concentration (MIC) of the ligand and its Cu(II) and Zn(II) complexes

Test Samples	MIC for different Pathogenic Bacterial Strains		
	<i>Staphylococcus aureus</i>	<i>Escherichia coli</i>	<i>Pseudomonas</i>
	ATCC 25926	ATCC 25922	<i>aeruginosa</i> ATCC 248
Ligand	25	25	25
CuL	12.5	12.5	12.5
CuL ₂	12.5	25	25
ZnL	25	12.5	12.5
ZnL ₂	25	12.5	12.5
Gentamycin	1.688	1.688	3.375
DMSO	ND	ND	ND

4.15.2.5. Antibacterial activities decorated AgNP-with ligand and its Cu(II) and Zn(II) complexes

Three microorganisms representing different microbial categories: one gram-positive (*S. aureus*) and two gram-negative (*E. coli* and *P. aeruginosa*)-have been used to study the

biological activity of the synthesized compounds. The results are summarized in Table 19, Appendix 14 and Appendix 15. The results are summarized in Table 19 that the metal complexes decorated with AgNPs show increased antibacterial activities than their undecorated complexes and bare AgNPs. Decorated AgNPs with the synthesized ligand and the metal complexes showed increased antibacterial ligand activity. In particular, the AgNP@CuL indicated the highest antibacterial value with values of 12.00 ± 1.00 , 10 ± 1.73 , and 11.33 ± 1.53 , against *S. aureus*, *E. coli*, and *P. aeruginosa*, respectively. The combination of AgNPs with bioactive compounds shows improved efficacy in specific treatments by lowering the side effects associated with the bioactive molecules. The small size of AgNPs could probably play a role in blocking cell communication mechanisms.

Table 19. Antimicrobial properties of the decorated ligand and metal complexes with AgNPs

Molecules (50 $\mu\text{g/mL}$)	Zone inhibition diameter (mm)		
	Gram-Positive bacterial		Gram-Negative bacterial
	<i>S. aureus</i>	<i>E. coli</i>	<i>P. aeruginosa</i>
AgNP@L	9.67 ± 0.58	9 ± 1.00	10.33 ± 0.58
AgNP@CuL	12 ± 1.00	10 ± 1.73	11.33 ± 1.53
AgNP@CuL ₂	8 ± 1.00	8.33 ± 0.57	8.67 ± 0.76
AgNP@ZnL	10.33 ± 0.01	9.77 ± 0.25	8.70 ± 0.26
AgNP @ZnL ₂	10.33 ± 0.76	9.47 ± 0.55	8.97 ± 1.00
Gentamycin (Positive control)	18.33 ± 0.58	17 ± 0	17.66 ± 0.58
DMSO (Negative control)	ND	ND	DD

4.15.2.6. Minimum Inhibitory Concentration of decorated AgNPs with the ligand, and its Cu(II) and Zn(II) Complexes

The minimum inhibitory concentration (MIC) of decorated AgNPs with imine quinoline ligand and its Cu(II) and Zn(II) complexes were evaluated against three bacterial pathogens and the results were tabulated in (Table 20). In the case of AgNP@L, the MIC was $12.5 \mu\text{g/mL}$ against both *E. coli* and *S. aureus*, while it was $25 \mu\text{g/mL}$ for *P. aeruginosa*. MICs of AgNP@CuL, AgNP@CuL₂, AgNP@ZnL and AgNP@ZnL were $12.5 \mu\text{g/mL}$ against *S.*

aureus, *E. coli* and *P. aeruginosa*. The MIC results confirm potent antibacterial activity against the three bacteria tested.

Table 20. The minimum inhibition concentration (MIC) of the decorated AgNP-with Cu(II) and Zn(II) complexes

Test Samples	MIC for different Pathogenic Bacterial Strains		
	<i>Staphylococcus aureus</i>	<i>Escherichia coli</i>	<i>Pseudomonas</i>
	ATCC 25926	ATCC 25922	<i>aeruginosa</i> ATCC 248
AgNP@ L	12.5	12.5	25
AgNP@ CuL	12.5	12.5	12.5
AgNP@CuL₂	12.5	12.5	12.5
AgNP@ZnL	12.5	12.5	12.5
AgNP@ZnL₂	12.5	12.5	12.5
Gentamycin (Positive)	1.688	1.688	3.375
DMSO (Negative)	ND	ND	ND

4.15.3. Antioxidant Activities

4.15.3.1. Antioxidant studies of biogenic synthesized and functionalized AgNPs

The antioxidant activity of compounds tested by DPPH methods. The green synthesized AgNP, AgNP-His, and AgNP-phen have showed DPPH free radical scavenging activity (antioxidant properties). The maximum scavenging activity was for AgNP-His (63.76±1.25%) at a concentration of 250 µg/mL followed by AgNP-phen and AgNP with 54.32±1.21% and 50.29±1.79%, respectively, at the same concentration as shown in Figure 39 and Appendix 16. The results also revealed that the antioxidant activity of the biogenic synthesized AgNP, AgNP-his, and AgNP-phen increased with an increasing concentration of test nanoparticle. This enhanced activity may be attributed due to functionalized of AgNPs with histidine and phenylalanine that provide stability and more sensitive scavenger activity of the nanoparticle (Kiymaci *et al.*, 2022). The sensitivity of the nanoparticles in antioxidant activity decreases in the order of AA.> AgNP-His > AgNP-Phen > AgNPs. The half inhibition concentration to

scavenging activity was calculated that AgNP > AgNP-Phen > AgNP-His with values of 251.9±0.85, 209.0±0.87, 160.4±1.030 µg/mL, respectively (Table 21).

Table 21. The half inhibition concentration to scavenging activity the AgNPs

	AgNP.Scv	AgNP-H.Scv	AgNP-Ph.Scv	AA.Scv
IC₅₀	251.9±0.85	160.4±1.03	209.0±0.88	1.034±0.72
logIC₅₀	2.401	2.205	2.320	0.01451

4.15.3.2. Antioxidant activity of the ligand and Zn(II) and Cu(II) metal complexes

The antioxidant activities of the imine quinoline ligand and its Zn(II) and Cu(II) complexes were investigated. It is crucial to research a compound's ability to scavenge free radicals since doing so may help stop the damaging impact that free radicals (Adeleke *et al.*, 2021; Chaudhary *et al.*, 2023). Consequently, we investigated the complexes capacity to scavenge 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radicals in vitro for the activities, using ascorbic acid as the standard. The complexes' ability to scavenge free radicals may be assessed by observing a reduction in the absorbance value of DPPH at 517 nm. Scavenging activity rises with increasing concentrations of the synthesized compounds that at high concentrations, high percentage of scavenging activities was recorded as shown in Figure 40 and Appendix 17.

The antioxidant activities of the imine-quinoline ligand and Cu(II) complexes were also assessed. The complexes' capacity to scavenge 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radicals were evaluated in vitro using ascorbic acid as the standard. The complexes' ability to scavenge free radicals was assessed by observing a reduction in the absorbance value of DPPH at 517 nm. Scavenging activity rises with increasing concentrations of complexes as shown in Figure 40. When compared to the reference ascorbic acid, all the complexes showed moderate activity. The metal complex of CuL has higher antioxidant ability than the metal complexes and imine quinoline ligand. Similarly, the half inhibition concentration (IC₅₀) was calculated that L > ZnL₂ > CuL₂ > ZnL > CuL with the values of 239.0±0.83, 198.3±0.86, 192.5±0.86, 167.0±1.04, and 153.3±1.02 µg/mL respectively (Table 22).

Table 22. The half inhibition concentration to scavenging activity the ligand and Cu(II) and Zn(II) complexes

	Ligand	CuL.Scv	CuL ₂ .Scv	ZnL.Scv	ZnL ₂ .Scv	AA.Scv
IC₅₀	239.0±0.83	153.3±1.02	192.5±0.86	167.0±1.04	198.3±0.86	1.034±0.72
logIC₅₀	2.378	2.186	2.285	2.223	2.297	0.01451

4.15.3.3. Antioxidant activities of decorated AgNP with ligand and metal complexes

The antioxidant activities of the synthesized decorated AgNP with ligand and its Zn(II) Cu(II) complexes were investigated. The decorated nanoparticles' capacity to scavenge 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radicals in vitro was evaluated using ascorbic acid as the standard. The capability to scavenge free radicals was assessed by observing a reduction in the absorbance value of DPPH at 517 nm. Scavenging activity rises with increasing concentrations of the decorated AgNPs: His-AgNP@CuL > His-AgNP@ZnL > His-AgNP@ZnL₂ > His-AgNP@CuL₂ > His-AgNP@L, with a value of (72.62±0.72, 67.58±1.67, 58.05±.90, 57.73±0.01, and 54.69±0.27) at 250 µg/mL, respectively, shown in Figure 41 and Appendix 18. The IC₅₀ of His-AgNP@CuL < His-AgNP@ZnL < His-AgNP@CuL₂ < His-AgNP@ZnL₂ and His-AgNP@L are 105.9± 0.97, 137.6±1.02, 184.1±0.85, 191.3±0.84, and 215.7±0.81 respectively, as shown in Table 23 Compared to the standard, ascorbic acid, all of the complexes showed moderate activity, as seen from the results.

Table 23 . The half inhibition concentration to scavenging activity of the decorated ligand and Cu(II) and Zn(II) complexes with AgNP

	His-AgNP@L	His-AgNP@CuL	His-AgNP@CuL ₂	His-AgNP@ZnL	His-AgNP@ZnL ₂	AA.Scv
IC₅₀	215.7±0.81	105.9±0.98	184.1±0.85	137.6±1.02	191.3±0.84	1.034±0.7

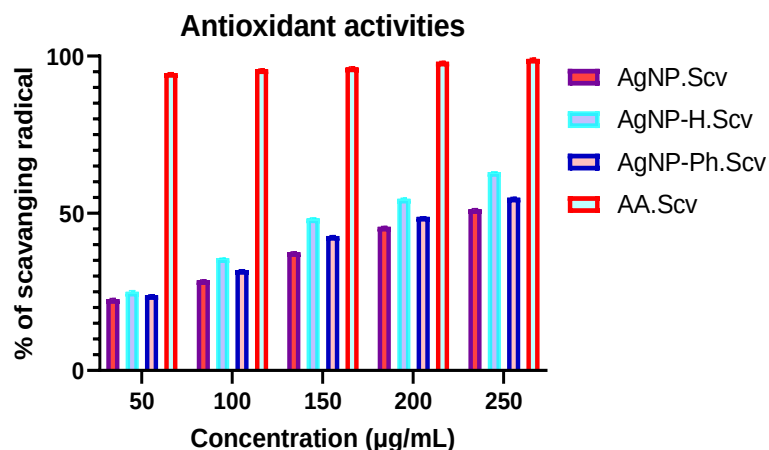


Figure 39. The antioxidant activity as scavenge percentage of biogenic synthesized AgNP, AgNP-His and AgNP-Phen using DPPH.

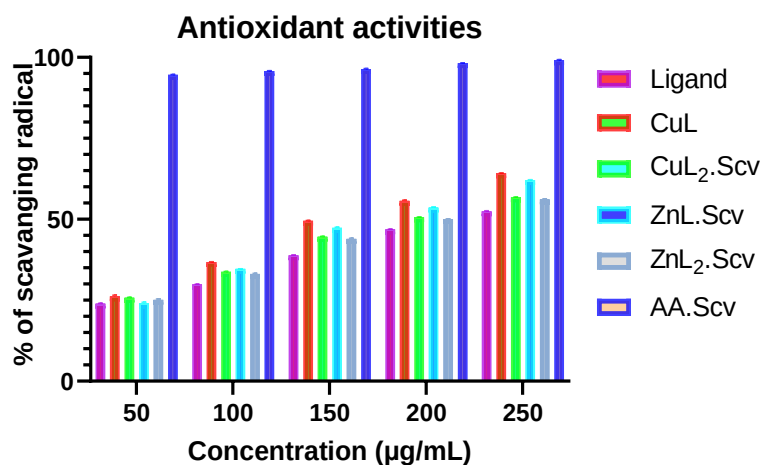


Figure 40. The antioxidant activities of the ligand (L) and its Cu and Zn ion metal complex

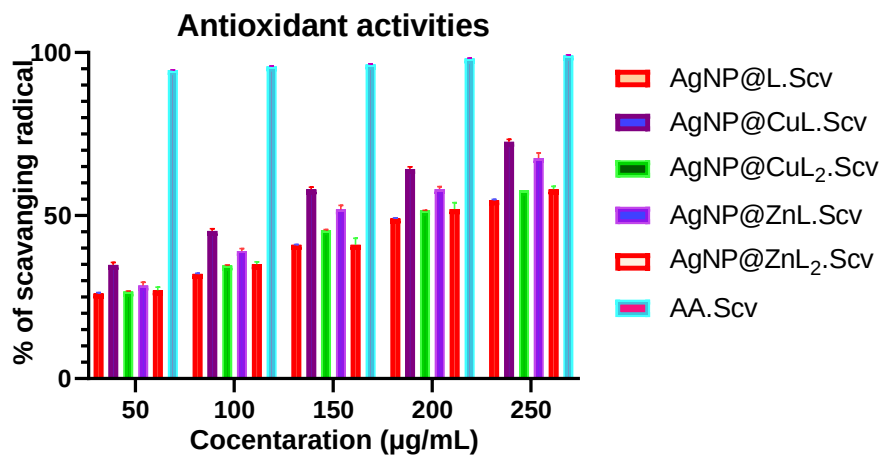


Figure 41. The antioxidant activities of the decorated ligand and Cu(II) and Zn(II) complexes with AgNP

4.15.4. Molecular Docking

4.15.4.1. Molecular Docking Analysis of zinc complexes

The 3D and 2D interaction of the synthesized imine quinoline ligand and the zinc complexes with *S. aureus* (PDB: 2w9h) and *E. coli* (PDB: 6F86) residual amino acid along with binding energy (kcal/mol) and inhibition constant is shown in Table 24. The imine quinoline ligand interaction with *S. aureus* (PDB: 2w9h) residual amino acid showed binding energy and inhibition constant values of -11.31 kcal/mol and 0.05 μM respectively. There are a total of 15 amino acid interaction with different binding modes includes 2 with hydrogen bond, 9 with van der Waals, 2 with π -alkyl and one with π -sigma as shown in Figure 42. The **ZnL** metal complex interaction with *S. aureus* (PDB: 2w9h) residual amino acid exhibited binding energy and inhibition constant values of -11.38 and 0.05 μM respectively with 10 of the residual amino acid interaction through different binding modes. These are 4 with hydrogen bond, 4 with van der Waals, one with π -alkyl and one with π -sigma as shown in Figure 43. **ZnL₂** metal complex interaction with *S. aureus* (PDB: 2w9h) residual amino acid showed binding energy and inhibition constant values -13.51 kcal/mol and 1.2×10^{-4} μM respectively with a total of 12 the residual amino acid interaction through different binding modes. These are 3 with hydrogen bond, 6 with van der Waals, one with π -alkyl, and 2 with π -sigma as shown in Figure 44. The gentamicin was used as positive control that the interaction with *S. aureus* (PDB: 2w9h) residual amino acid exhibited binding energy and inhibition constant value of -10.78 kcal/mol and 0.13 μM , respectively with a maximum of 21 the residual amino acid interaction through different binding modes without interaction through π -sigma as shown in Figure 45. Similarly, the imine quinoline ligand, **ZnL** and **ZnL₂** complexes interaction with *E. coli* (PDB: 6F86) residual amino acid were computed and the data tabulated in Table 25 that imine-quinoline ligand showed binding energy and inhibition constant values of -7.92 kcal/mol and 1.57 μM , respectively. There are total of 16 amino acid interaction with different binding modes namely 2 with hydrogen bond, 9 with van der Waals, 4 with π -alkyl and one with π -sigma as shown in Figure 46. The **ZnL** complex was interacted with *E. coli* (PDB: 6F86) residual amino acid perceived binding energy and inhibition constant values of -8.07 kcal/mol and 1.22 μM , respectively a total of 15 residual amino acid interaction through

different binding modes namely 2 with hydrogen bond, 12 with van der Waals, one with π -alkyl and no interaction of π -sigma as shown in Figure 47. **ZnL₂** metal complex interaction with *E. coli* (PDB: 6F86) residual amino acid perceived binding energy and inhibition constant value of -8.92 kcal/mol and 0.29 μ M, respectively with a total of 17 residual amino acid interaction through different binding modes. These are 2 with Hydrogen bond, 6 with van der Waals, 8 with π -alkyl, and one with π -sigma as shown in Figure 48. The gentamicin was used as positive control that the interaction with *E. coli* (PDB: 6F86) residual amino acid perceived binding energy and inhibition constant value of -10.10 kcal/mol and 39.31 μ M respectively with a maximum a total of 16 the residual amino acid interaction through different binding modes without interaction through π -sigma as shown in Figure 49.

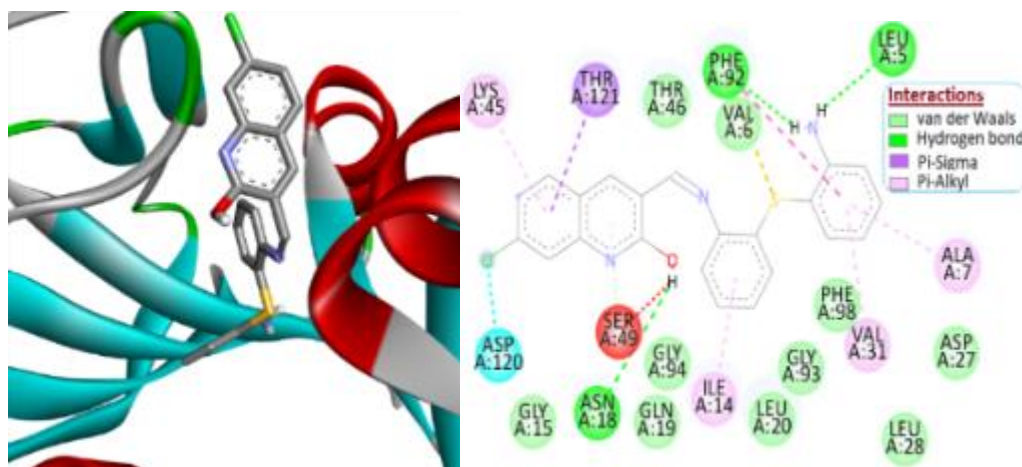


Figure 42. The binding interactions of imine quinoline ligand (L) against *S. aureus* (PDB: 2w9h)

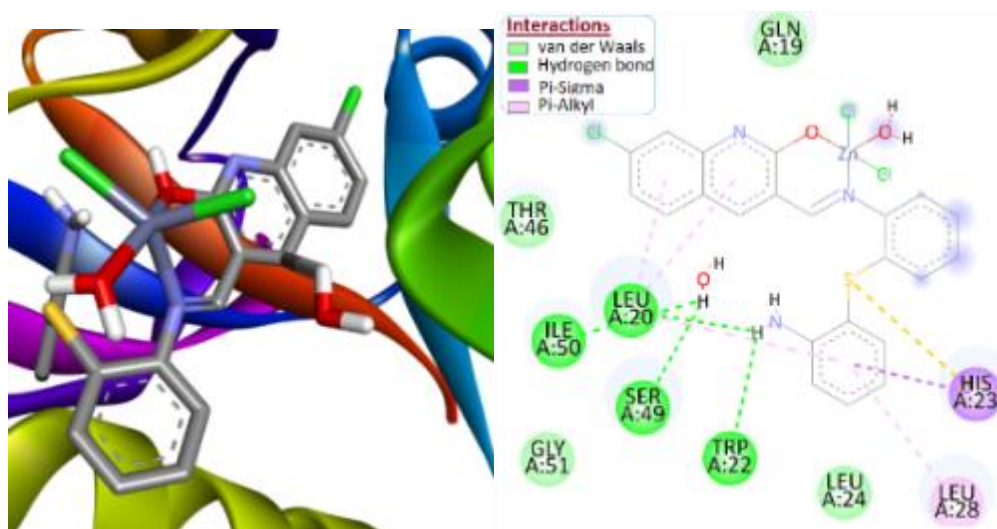


Figure 43. The binding interactions of **ZnL** complex against *S. aureus* (PDB: 2w9h)

Table 24. Molecular docking scores and the corresponding prominent residual amino acid interactions of the ligand and its metal complexes against *S. aureus* (PDB: 2w9h)

Cpds	Binding energy (kcal/mol)	Inhibition constant (K_i)	H-bonding	van der Waals	π -alkyl	π Sigma
L	-11.31	0.05 μ M	PHE 92, Leu 5, Asn 18	Thr 46, Val 6, Phe 98, Asp 27, Leu 28, Leu 20, Gly 93, Gln 19, Gln15	Lys 45, Ala 7,	Thr 121
ZnL	-11.38	0.05 μ M	Leu 28, Ile 50, Ser 49, Trp 22,	Gln 19, Thr 46, Gly 51, Leu 24	His 23	Leu 28
ZnL₂	-13.51	1.2×10^{-4} μ M	Asn 18, Ser 49, Ile 50	Gln 19, His 23, Thr 46, Phe 92, Gly 51, Phe 92	Leu 20	Leu 28, Leu 54
Gentamicin	-10.78	0.13 μ M	Ile 14, Ser 49, Asp 27, Phe 92	Leu 54, Thr 46, Thr 121, Gly 94, Gly 93, Gly 15, Phe 98, Phe 16, Leu 28, Val 31, Asn 18, Gln 95, 19	Ile 50, Leu 20, Val 6, Ala 7, lys 45	

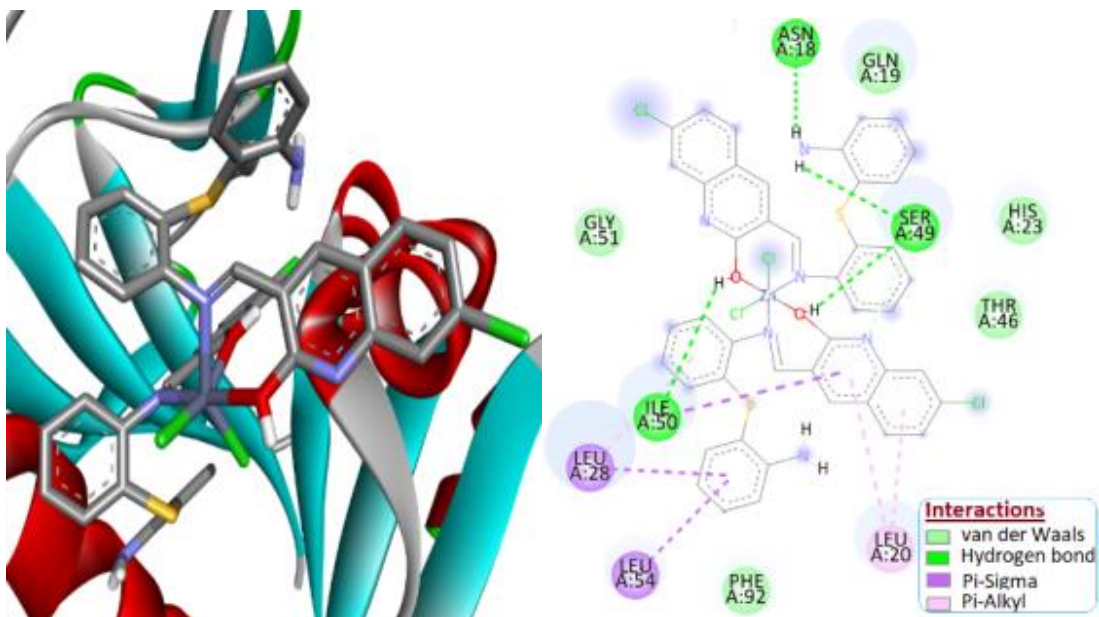


Figure 44. The binding interactions of **ZnL₂** complex against *S. aureus* (PDB: 2w9h)

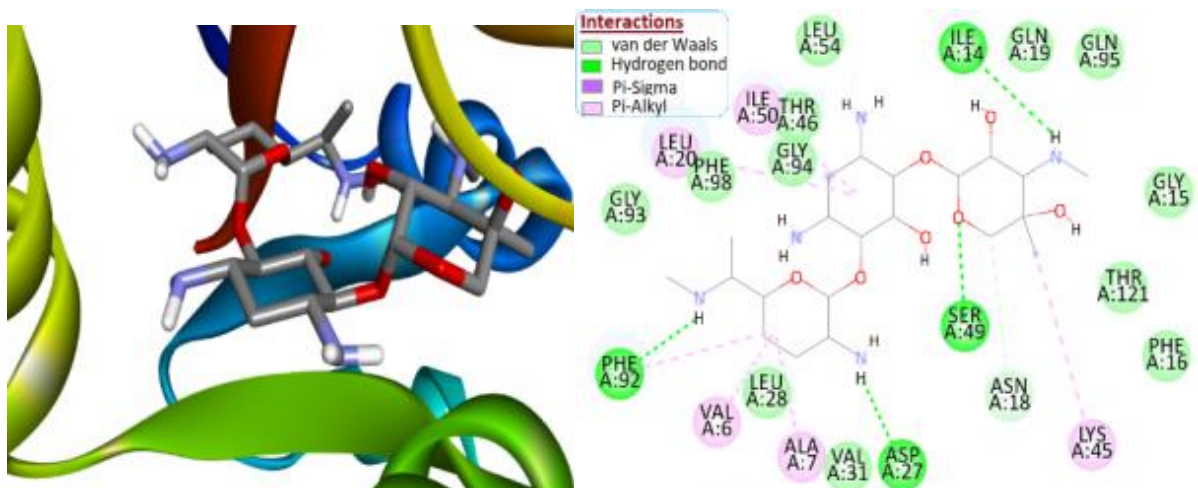


Figure 45. The binding interactions of gentamicin against *S. aureus* (PDB: 2w9h)

Table 25. Molecular docking scores and the corresponding prominent residual amino acid interactions of the ligand and its metal complexes against *E. coli* (PDB: 6F86)

Cpds	Binding energy (kcal/mol)	Inhibition constant (K_i)	H-bonding	van der Waals	π -alkyl	Pi-Sigma
L	-7.92	1.57 μ M	Arg 76, Glu 50	Ile 94, Pro 79, Asn 46, Val 120, Val 43, Asp 73, Asp 49, Gly 77, Gly 75	Ala 53, Ala 47, Val 167, Ile 78	Thr 165
ZnL	-8.07	1.22 μ M	ASN 46, ASP 73	Val 118, Val 120, Ser121, Leu 98, Gly 119, Gly 75, Gly 77, Ile 94, Pro 79, Ala 47, Thr 165, Arg 76	Ile 78	
ZnL₂	-8.92	0.29 μ M	Leu 98, Asn 46	Asp 45, 73, Val 120, Val 93, Val 71, Gly 77	Pro 79, Arg76, Ala 47, Ile 78, 94, Val 43, Val 167, Val 97	Thr 165
Gentamicin	-10.10	39.31 μ M	Ile 50, Ser 49, Asp 27, Phe 92	Gln 19, Leu 28, Leu 5, Ala 7, Thr 46, Val 31, Gly 94	Leu 54, Leu 20, Ile 14, Val 6, Phe 98	

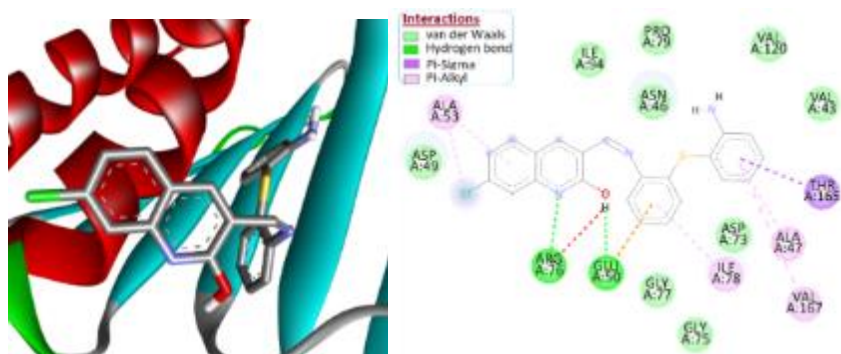


Figure 46. The binding interactions of imine quinoline ligand(L) against *E. coli* (PDB: 6F86)

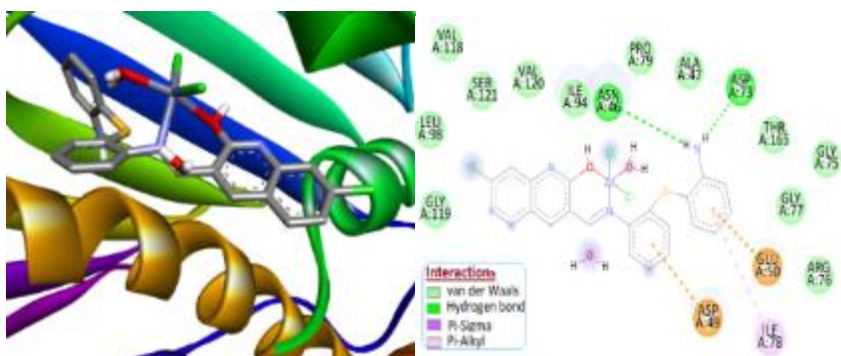


Figure 47. The binding interactions of **ZnL** against *E. coli* (PDB: 6F86)

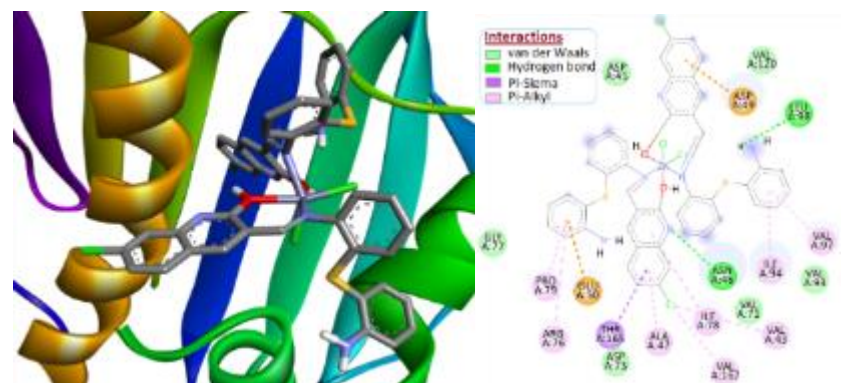


Figure 48. The binding interactions of **ZnL₂** against *E. coli* (PDB: 6F86)

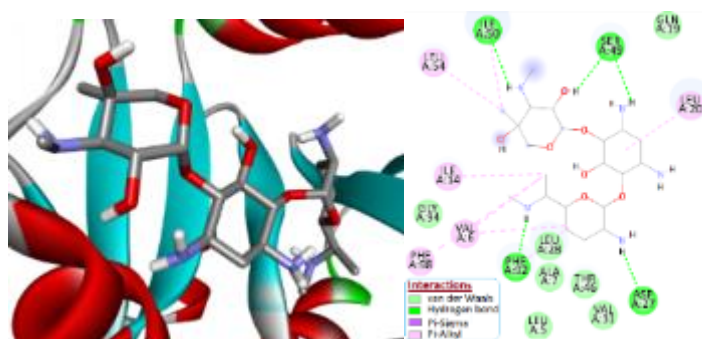


Figure 49. The binding interactions of gentamicin against *E. coli* (PDB: 6F86)

4.15.4.2. Molecular Docking copper complexes

Table 26 - 27 and Figure 50 - 57 present the 3D and 2D interactions of the synthesized **L** and the Cu(II) complexes with residual amino acids *S. aureus*, *E. coli*, and *ERα*, along with their respective binding energies and inhibition constants. The interaction of **L** with residual amino acids from *S. aureus* resulted in a binding energy of -11.31 kcal/mol and an inhibition constant of 0.05 μ M. A total of 15 amino acid interactions were identified with different binding modes. 2 via hydrogen bonds, 9 via van der Waals forces, 2 via π -alkyl interactions, and 1 via π -sigma interaction, as depicted in Figure 50.

The interaction of the **CuL** with residual amino acids of *S. aureus* was evaluated, revealing a binding energy of -13.82 kcal/mol and an inhibition constant of 7.5×10^{-5} μ M. A total of 18 residual amino acid interactions were identified, that occurs through various binding modes: 4 via hydrogen bonds, 10 via van der Waals forces, 3 via π -alkyl interactions, and 1 via π -sigma interaction as shown in Figure 51. Similarly, **CuL₂** metal complex exhibited interaction with *S. aureus* residual amino acids, yielding a binding energy of -12.19 kcal/mol and an inhibition constant of 0.01 μ M. A total of 8 residual amino acid interactions were observed that involves different binding modes: 1 via hydrogen bond, 1 via van der Waals forces, 3 via π -alkyl interactions, and 3 via π -sigma interactions (Figure 52). Gentamicin served as a positive control, displaying a binding energy of -10.78 kcal/mol and an inhibition constant of 0.13 μ M. A total of 21 residual amino acid interactions were noted, with various binding modes, excluding interactions via π -sigma shown in Figure 53.

Furthermore, the interactions of **L**, **CuL**, and **CuL₂** with residual amino acids of *E. coli* were analyzed. **L** demonstrated a binding energy of -7.92 kcal/mol and an inhibition constant of 1.57 μ M. Sixteen amino acid interactions were identified with various binding modes: 2 via hydrogen bonds, 9 via van der Waals forces, 4 via π -alkyl interactions, and 1 via π -sigma interaction (Figure 54). **CuL** interacted with *E. coli* residual amino acids, displaying a binding energy of -7.98 kcal/mol and an inhibition constant of 1.41 μ M. Fifteen residual amino acid interactions were observed, occurring through different binding modes: 1 via hydrogen bond, 6 via van der Waals forces, 5 via π -alkyl interactions, and 3 via π -sigma interactions showed in

Figure 55. Similarly, **CuL₂** interacted with *E. coli* residual amino acids, showing a binding energy of -8.92 kcal/mol and an inhibition constant of 0.29 μ M. Eleven residual amino acid interactions were noted, with different binding modes: 3 via hydrogen bonds, 4 via van der Waals forces, 3 via π -alkyl interactions, and one via π -sigma interaction (Figure 56). Gentamicin was utilized as a positive control for interaction with *E. coli* residual amino acids, exhibiting a binding energy of -10.10 kcal/mol and an inhibition constant of 39.31 μ M. Sixteen residual amino acid interactions were identified, involving various binding modes, except for interactions via π -sigma (Figure 57).

The docking of **L**, **CuL**, **CuL₂**, and Cisplatin against estrogen receptor alpha were assessed, are given in Table 28 and Figures 58-60. The interaction of **L** with *ER α* residual amino acids revealed a binding energy of -7.63 kcal/mol and an inhibition constant of 2.53 μ M. A total of 14 amino acid interactions were identified, involving different binding modes: 2 through hydrogen bonds, 10 through van der Waals forces, and 2 through π -alkyl interactions showed in Figure 58. **CuL** displayed a binding energy of -8.79 kcal/mol and an inhibition constant of 0.36 μ M, with a total of 13 amino acid interactions occurring through various binding modes, including 3 through hydrogen bonds, 5 through van der Waals forces, 1 through π -alkyl, 3 through π - π , and 1 through π -anion interactions (Figure 59). Similarly, **CuL₂** exhibited a binding energy of -8.66 kcal/mol and an inhibition constant of 0.45 μ M, with a total of 14 amino acid interactions occurring through different binding modes: 3 through hydrogen bonds, 9 through van der Waals forces, 1 through π -alkyl, and 2 through π -anion interactions (Figure 60). Cisplatin demonstrated a binding energy and inhibition constant of -6.09 kcal/mol and 34.40 μ M, respectively.

Table 26. Molecular docking scores and the corresponding prominent residual amino acid interactions of the ligand and its Cu(II) complexes against *S. aureus* (PDB: 2w9h)

Cpds	Binding energy (kcal/mol)	Inhibition constant (K_i)	H-bonding	van der Waals	π -alkyl	Pi Sigma
L	-11.31	0.05 μ M	Phe 92, Leu 5, Asn 18	Thr 46, Val 6, Phe 98, Asp 27, Leu 28 Leu 20, Gly 93, Gln 19, Gln 15	Lys45, Ala 7,	Thr 121
CuL	-13.82	7.5×10^{-5} μ M	Phe 92, Leu 5, Asn 18, Ile 14	Val 6, Asp 27, Phe 98, Gly 93, Gly 15, Gly 94, Thr 121, Thr 46, Gln 95, Gln 19	Val 31, Ala 7, Leu 20	Ile 50
CuL₂	-12.19	0.01 μ M	Ser 49	Gln 19	Val 31, Pro 25, Leu 20	Leu 28, Leu 54, Ile 50
Gentamicin	-10.78	0.13 μ M	Ile 14, Ser 49, Asp 27, Phe 92	Leu 54, Thr 46, Thr 121 Gly 94, Gly 93, Gly 15, Phe 98, 16, Leu 28, Val 31, Asn 18, Gln 95, Gln 19	Ile 50, Leu 20, Val 6, Ala 7, Lys 45	



Figure 50. The binding interactions of L against *S. aureus* (PDB: 2w9h)

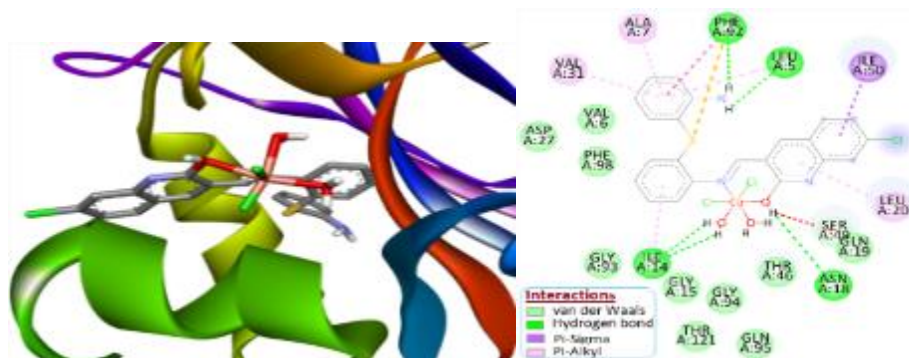


Figure 51. The binding interactions of CuL against *S. aureus* (PDB: 2w9h)

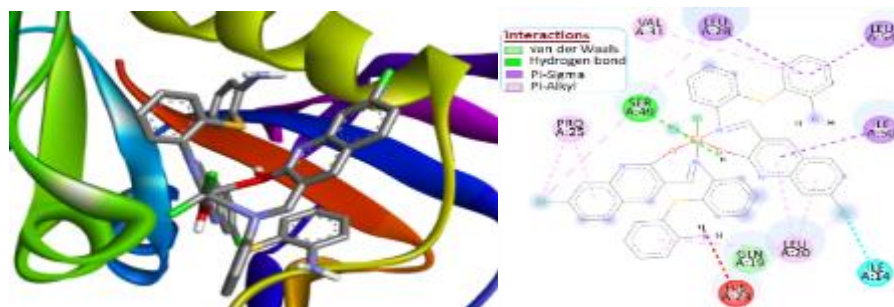


Figure 52. The binding interactions of CuL₂ against *S. aureus* (PDB: 2w9h)

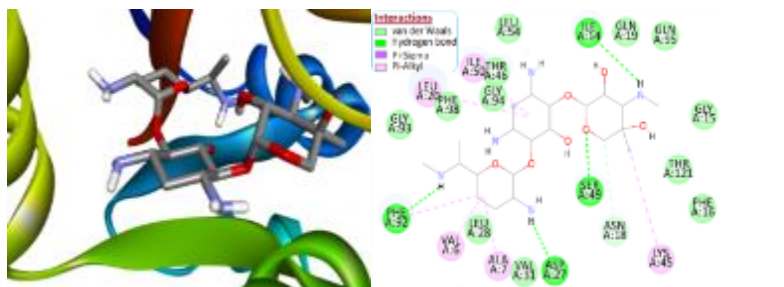


Figure 53. The binding interactions of gentamicin against *S. aureus* (PDB: 2w9h)

Table 27. Molecular docking scores and the corresponding prominent residual amino acid interactions of the ligand and its Cu(II) complexes against *E. coli* (PDB: 6F86)

Cpds	Binding energy (kcal/mol)	Inhibition constant (K_i)	H-bonding	van der Waals	π -alkyl	Pi-Sigma
L	-7.92	1.57 μ M	Arg 76, Glu 50	Ile94, Pro 79, Asn 46, Val 120, Val 43, Asp 73,49, Gly 77,Gly 75	Ala 53,Ala 47, Val 167, Ile 78	Thr 165
CuL	-7.98	1.41 μ M	Asn 46	Asp 73, Asp 49, Glu 50, Pro 79, Val 167, Gly 119	Ala 47, Ala 43, Val 71, Val 120, Val 43	Ile 78, Ile94, Thr 165
CuL₂	-7.37	3.95 μ M	Asn 46, Ile 94, Val 93	Ala 53, Gly 117, Gly 119, Glu 42, Ser 121, Thr 165	Ile 78, Arg 76, Val 120	Gly 77
Gentamicin	-10.10	39.31 μ M	Ile 50, Ser 49, Asp 27, Phe92	Gln 19, Leu 28, Leu 5, Ala 7, Thr 46, Val 31, Gly 94	Leu 54, Leu 20, Ile 14, Val 6, Phe 98	

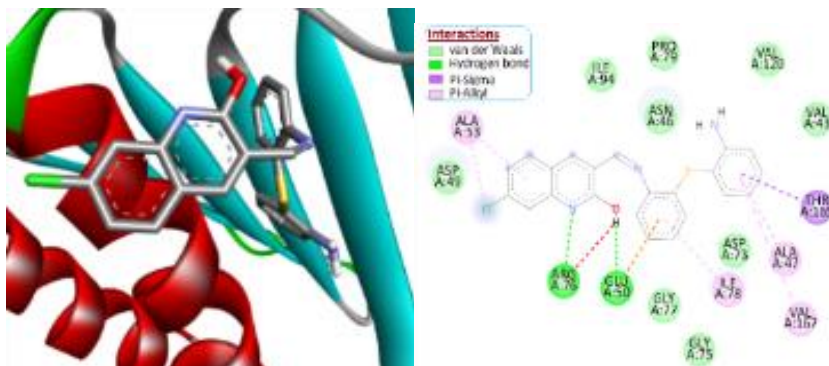


Figure 54. The binding interactions of L against *E. coli* (PDB: 6F86)

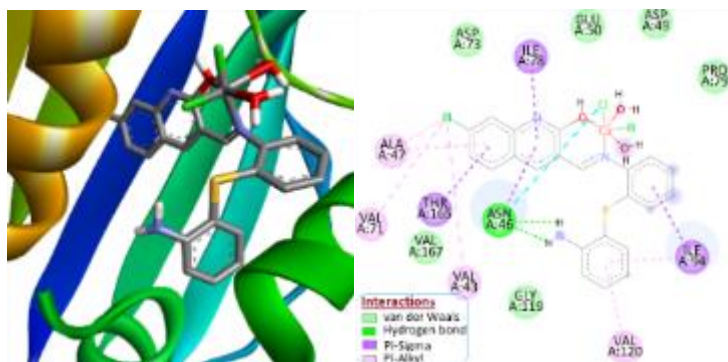


Figure 55. The binding interactions of CuL against *E. coli* (PDB: 6F86)

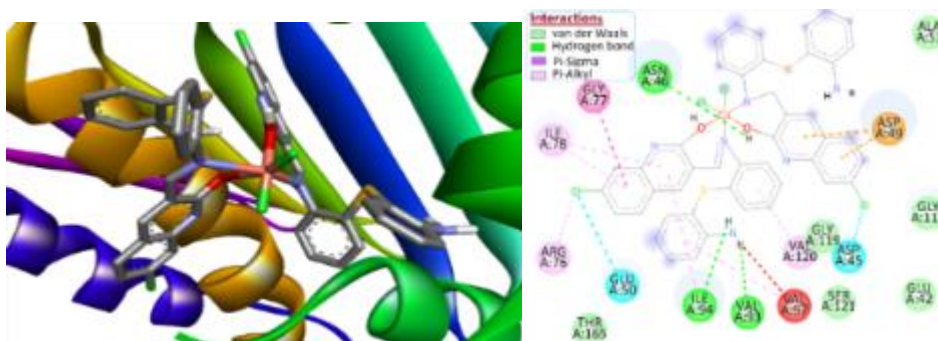


Figure 56. The binding interactions of CuL₂ against *E. coli* (PDB: 6F86)

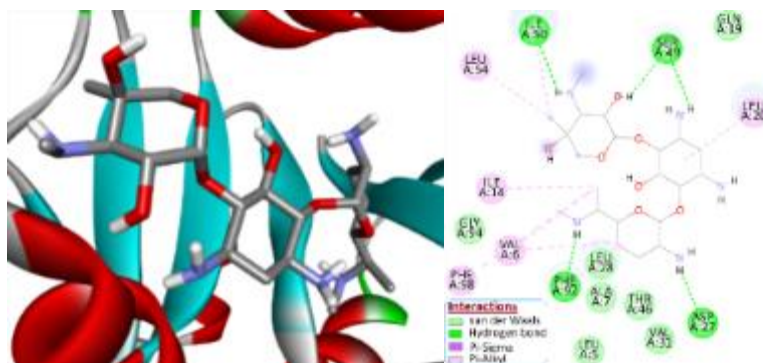


Figure 57. The binding interactions of gentamicin against *E. coli* (PDB: 6F86)

Table 28. Molecular docking scores and the corresponding residual amino acid interactions of the ligand and its Cu(II) complexes against estrogen receptor alpha (ER α ; PDB:5GS4)

Cpds	Binding energy (kcal/mol)	Inhibition constant (K _i)	H-bonding	van der Waals	π -alkyl	π - π	π -anion
IQL	-7.63	2.53 μ M	Glu353, Glu323	Arg394,Ala322, Asn359,Arg363, Gly390,Ile386, Leu387,Lys449, Phe445,Trp360,	Met357, His356		
CuL	-8.79	0.36 μ M	Arg394, Ile386, Glu353	Glu323,His356, Leu387,Gly390, Leu391	Met357	Phe445, Trp393, Trp360	Lys449
CuL₂	-8.66	0.45 μ M	Glu323, Asp 321	Pro406,Gly390, Leu387,Trp393, Phe445,Arg363, Ala322, Trp360	Met357		Arg394
Cisplatin	-6.09	34.40 μ M	Glu471,Asp 374, Ser468	Thr371	Lys467		

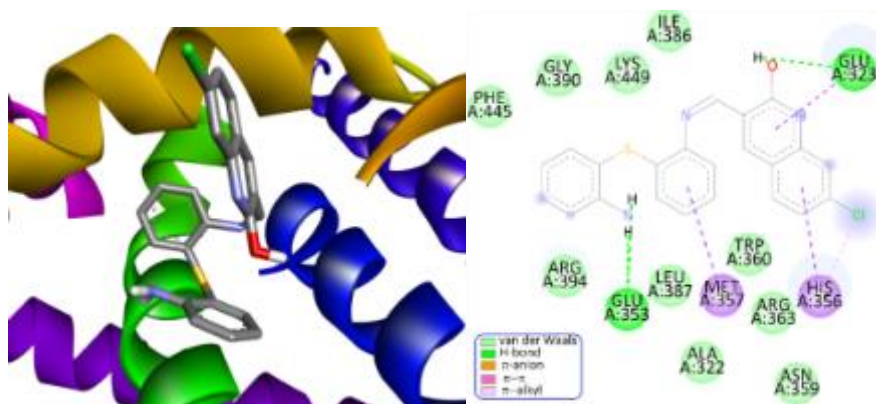


Figure 58. The-binding interactions of L against estrogen receptor alpha ($ER\alpha$; PDB: 5GS4)

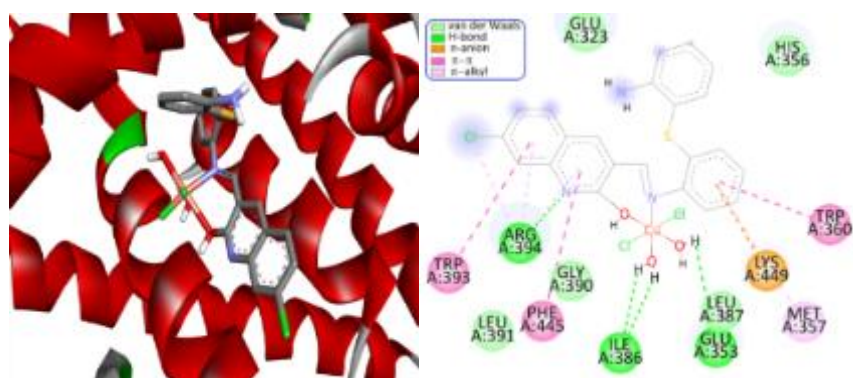


Figure 59. The binding interactions of **CuL** against estrogen receptor alpha ($ER\alpha$; PDB: 5GS4)

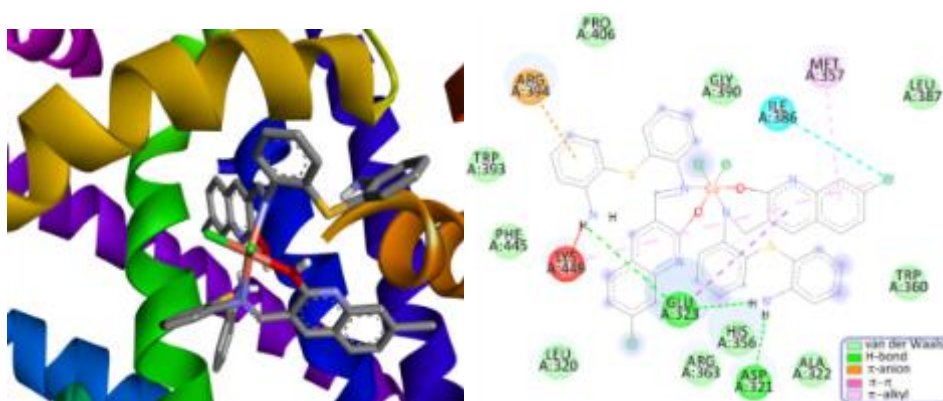


Figure 60. The binding interactions of **CuL₂** against estrogen receptor alpha ($ER\alpha$; PDB: 5GS4)

5. CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

This study was focused on the synthesis and characterization of the imine quinoline derivative ligand and its Cu(II) and Zn(II) complexes, $[\text{CuL}(\text{H}_2\text{O})\text{Cl}_2]$, $[\text{CuL}_2\text{Cl}_2]$, $[\text{ZnL}(\text{H}_2\text{O})\text{Cl}_2]$ and $[\text{ZnL}_2\text{Cl}_2]$ and their decorated biogenic of AgNPs (His-AgNP@L, His-AgNP@CuL, His-AgNP@CuL₂, His-AgNP@ZnL and His-AgNP@ZnL₂) with the aim of developing potential therapeutic agents with antimicrobial, and antioxidant properties. In addition, the anticancer activity of the imine quinoline ligand and its Cu(II) complexes were carried out against MCF-7 cancer lines. The characterization was conducted using various analytical techniques, including FTIR, pXRD, TGA/DTA, UV-Vis, TD-TDF, TEM, EDX-SEM molar conductivity, and chloride tests. The synthesized Cu(II) and Zn(II) complexes demonstrated thermal stability up to 200 °C, with pXRD confirming their nanocrystalline nature. The UV-Vis analysis supports the distinctive peak obtained for the synthesized AgNPs in the 400 – 450 nm range, and intra-ligand electron transfer, d-d electron transitions, and ligand-to-metal charge transfer for the ligand and the metals complexes. FTIR confirmed the functional groups of the nanoparticle, ligand, and complexes, with the complexes found to be non-electrolytic and chloride-free outside the coordination sphere, suggesting an octahedral geometry. TD-DFT absorption spectra validated and aligning well with experimental data for the synthesized ligand and the Cu(II) and Zn(II) complexes. Physicochemical analysis, spectrophotometric characterizations and TD-DFT computing confirmed the synthesis of ligand and its octahedral Cu(II) and Zn(II) complexes. Biological evaluations revealed that **AgNP@CuL** and CuL demonstrated strongest antioxidant and antibacterial activities compared to other synthesized materials with **CuL** exhibiting the comparable potent cytotoxicity, antibacterial, and antioxidant activities. The Zn(II) complexes exhibited better antibacterial and antioxidant activities than the imine-quinoline ligand. Frontier molecular orbital (FMO) analysis and molecular docking results were supporting the strong antioxidant and antibacterial activity of **CuL**. The molecular docking results revealed a higher binding affinity of the metal complexes to the binding sites of *S. aureus* compared to the ligand, which is consistent with the experimental findings. This suggests that the **CuL** complex may have potential therapeutic

applications against *S. aureus* infections. Therefore, it is recommended to conduct further *in vivo* studies to evaluate the efficacy, safety, and potential therapeutic benefits of the **CuL** and **AgNP@CuL** complex in treating *S. aureus* and *MCF-7* breast cancer infections.

5.2. Recommendations.

- ✚ In this study, biogenic synthesis and coordination chemistry were utilized as key strategies to synthesize nanoparticles using plant extracts and amino acids, along with metal complexes derived from bioactive organic ligands. The synthesized materials: silver nanoparticles (AgNPs), imine quinoline ligands, and their corresponding Cu(II) and Zn(II) metal complexes were extensively characterized using various techniques, including UV-Vis, FTIR, XRD, NMR, SEM-EDX, and TEM. Their *in vitro* antibacterial, anticancer, and antioxidant activities were also evaluated. The results demonstrated that AgNP-His, the imine quinoline ligand, and the metal complexes exhibited significant antibacterial and antioxidant properties, while the Cu(II) complex showed promising anticancer activity, further supported by *in silicon* studies. These findings suggest that such materials have the potential to address the growing challenge of antibiotic resistance and serve as an effective strategy for future therapeutic applications.
- ✚ While this study focused on concentrations above 2 mM, future studies could explore lower concentrations to determine the minimum silver ion concentration required for efficient nanoparticle production. This could help identify if lower concentrations provide adequate yield while reducing reagent costs and minimizing potential environmental impact.
- ✚ Characterizing the interactions between histidine-functionalized AgNPs and Cu(II) or Zn(II) complexes with imine-quinoline ligand was limited in this study, future studies should utilize advanced techniques. X-ray photoelectron spectroscopy (XPS) could confirm coordination bonding and ligand exchange, while electron paramagnetic resonance (EPR) can provide detailed insights into the binding environment and oxidation states. Dynamic light scattering (DLS) would be useful for real-time monitoring and detecting changes in nanoparticle size or aggregation. Additionally, computational modeling, such as density functional theory (DFT), could predict binding geometries and

stability. Electrochemical techniques like cyclic voltammetry could help understand redox interactions between AgNPs and the metal complexes. These approaches would allow for a more comprehensive understanding of the interactions and optimize the design of these functionalized nanoparticles for biological and catalytic applications.

- ✚ In vivo studies should be prioritized in future research to better understand the biological efficacy and safety of these materials under physiological conditions. These evaluations would provide crucial insights into their biocompatibility, toxicity, and real-world therapeutic potential, offering a more complete understanding of their performance for potential biomedical applications.
- ✚ Explore the synergistic potential between the synthesized AgNPs, imine ligands, and Cu(II) and Zn(II) complexes. Combining these materials could enhance their biological efficacy, particularly in cancer treatment and antimicrobial applications, while allowing you to use lower concentrations and minimize toxicity.

6. REFERENCES

- Abate, Chiara, Federica Carnamucio, Ottavia Giuffrè, and Claudia Foti. 2022. "Metal-Based Compounds in Antiviral Therapy." *Biomolecules* 12(7).
- Abbas, Abbas M., Hossam H. Nasrallah, Ahmed Aboelmagd, Safaa M. Kishk, W. Christopher Boyd, Haitham Kalil, and Adel S. Orabi. 2024. "Design, Synthesis, Anti-Inflammatory Activity, DFT Modeling and Docking Study of New Ibuprofen Derivatives." *International Journal of Molecular Sciences* 25(6):3558(1-22).
- Abd El-Lateef, Hany M., Mai M. Khalaf, Amer A. Amer, Mahmoud Kandeel, Antar A. Abdelhamid, and Aly Abdou. 2023. "Synthesis, Characterization, Antimicrobial, Density Functional Theory, and Molecular Docking Studies of Novel Mn(II), Fe(III), and Cr(III) Complexes Incorporating 4-(2-Hydroxyphenyl Azo)-1-Naphthol (Az)." *ACS Omega* 8(29):25877–91.
- Abdoon, Salwa Neama, Saba S. M. Al-Obaidy, and Yahya Fahim Al-Khafaji. 2022. "Synthesis, Spectral Analysis, Stability, Antibacterial, and Antioxidant of Fe(II) Mixed Ligands Complex of Imidazole and 1,10-Phenanthroline." *Journal of Medicinal and Chemical Sciences* 5(7):1231–41.
- Abu-Dief, Ahmed, Tarek El-dabea, Rafat El-Khatib, Aly Abdou, and Mahmoud Abd El Aleem El- Remailya. 2024. "Development of New Mixed Cu(II) Chelate Based on 2-Benzimidazolylguanidine and Phenanthroline Ligands: Structural Elucidation, Biological Evaluation, DFT and Docking Approaches." *Sohag Journal of Sciences* 9(2):174–85.
- Adeleke, Adesola A., Sizwe J. Zamisa, Md Shahidul Islam, Kolawole Olofinan, Veronica F. Salau, Chunderika Mocktar, and Bernard Omondi. 2021. "Quinoline Functionalized Schiff Base Silver (I) Complexes: Interactions with Biomolecules and in Vitro Cytotoxicity, Antioxidant and Antimicrobial Activities." *Molecules* 26(5).
- Agidew, Misganaw Gedlu. 2022. "Phytochemical Analysis of Some Selected Traditional Medicinal Plants in Ethiopia." *Bulletin of the National Research Centre* 46(1).
- Agu, P. C., C. A. Afiukwa, O. U. Orji, E. M. Ezech, I. H. Ofoke, C. O. Ogbu, E. I. Ugwuja, and P. M. Aja. 2023a. "Molecular Docking as a Tool for the Discovery of Molecular Targets of Nutraceuticals in Diseases Management." *Scientific Reports* 1–18.
- Agu, P. C., C. A. Afiukwa, O. U. Orji, E. M. Ezech, I. H. Ofoke, C. O. Ogbu, E. I. Ugwuja, and P. M. Aja. 2023b. "Molecular Docking as a Tool for the Discovery of Molecular Targets

- of Nutraceuticals in Diseases Management.” *Scientific Reports* 1–18.
- Ahmad, Faheem, Mounir M. Salem-Bekhit, Faryad Khan, Sultan Alshehri, Amir Khan, Mohammed M. Ghoneim, Hui Fen Wu, Ehab I. Taha, and Ibrahim Elbagory. 2022. “Unique Properties of Surface-Functionalized Nanoparticles for Bio-Application: Functionalization Mechanisms and Importance in Application.” *Nanomaterials* 12(8).
- Ahmad, Fawad, Nasir Abbas, Ayesha Ihsan, Muhammad Saqib Ghafoor, Ghulam Shabir, and Aamer Saeed. 2023. “Zn (II) Octahedral Complex with New Ketimine Ligand: Design, Synthesis and Single-Crystal Studies.” *Transition Metal Chemistry* 48(5):343–51.
- Ahmad, Zubair and Faiza Habib. 2023. “Metal-Organic Framework Based Nanomaterials : An Advanced Review of Their Synthesis and Energy Storage Applications.” (June).
- Ahmed, Samah M., Shaimaa Ali Abdelrahman, and Alaa Elsayed Salama. 2018. “Efficacy of Gold Nanoparticles against Isoproterenol Induced Acute Myocardial Infarction in Adult Male Albino Rats.” *Ultrastructural Pathology* 00(00):1–18.
- Ahmed, Sirwan Khalid, Safin Hussein, Karzan Qurbani, Radhwan Hussein Ibrahim, Abdulmalik Fareeq, Kochr Ali Mahmood, and Mona Gamal Mohamed. 2024. “Antimicrobial Resistance: Impacts, Challenges, and Future Prospects.” *Journal of Medicine, Surgery, and Public Health* 2(January):100081.
- Aishajiang, Reyida, Zhongshan Liu, Tiejun Wang, Liang Zhou, and Duo Yu. 2023. “Recent Advances in Cancer Therapeutic Copper-Based.” *Molecules* 28(2303):1–20.
- Ajani, Olayinka O., King T. Iyaye, and Olabisi T. Ademosun. 2022. “Recent Advances in Chemistry and Therapeutic Potential of Functionalized Quinoline Motifs - a Review.” *RSC Advances* 12(29):18594–614.
- Akbari, Zahra, Claudio Stagno, Nunzio Iraci, Thomas Efferth, Ejla A. Omer, Anna Piperno, Morteza Montazerzohori, Mehran Feizi-dehnayebi, and Nicola Micale. 2024. “Biological Evaluation , DFT , MEP , HOMO-LUMO Analysis and Ensemble Docking Studies of Zn (II) Complexes of Bidentate and Tetridentate Schiff Base Ligands as Antileukemia Agents.” *Journal of Molecular Structure* 1301(December 2023):137400.
- Akinpelu, Lateef Abiola, Olawuni Idowu, Gbenga Emmanuel Ogundepo, and Gbola Olayiwola. 2019. “Spectroscopic Analysis and Anti-Inflammatory Effects of *Milicia Excelsa* (Moraceae) Leaf and Fractions GSC Biological and Pharmaceutical Sciences Spectroscopic Analysis and Anti-Inflammatory Effects of *Milicia Excelsa* (Moraceae)

- Leaf and Fractions.” *GSC Biological and Pharmaceutical Sciences* 6(3):051–061.
- Al-Amiery, Ahmed A., Yasmien K. Al-Majedy, Heba H. Ibrahim, and Ali A. Al-Tamimi. 2012. “Antioxidant, Antimicrobial, and Theoretical Studies of the Thiosemicarbazone Derivative Schiff Base 2-(2-Imino-1-Methylimidazolidin-4-Ylidene)Hydrazinecarbothioamide (IMHC).” *Organic and Medicinal Chemistry Letters* 2(1):4.
- Al-daffay, Rasha Khider Hussain. 2023. “Synthesis , Characterization , and Thermal Analysis of a New Acidicazo Ligand ’ s Metal Complexes Abstract :” *Baghdad Science Journal* 20(July 2022):121–33.
- Al-Harbi, Nuha and Nabil K. Abd-Elrahman. 2024. “Physical Methods for Preparation of Nanomaterials, Their Characterization and Applications: A Review.” *Journal of Umm Al-Qura University for Applied Sciences* (0123456789).
- Al-Ostoot, Fares Hezam, Zabiulla, Salma Salah, and Shaukath Ara Khanum. 2021. *Recent Investigations into Synthesis and Pharmacological Activities of Phenoxy Acetamide and Its Derivatives (Chalcone, Indole and Quinoline) as Possible Therapeutic Candidates*. Vol. 18. Springer Berlin Heidelberg.
- Al-Qadsy, Inas, Waseem Sharaf Saeed, Abdel Basit Al-Odayni, Ali Alrabie, Lena Ahmed Saleh Al-Faqeeh, Arwa Al-Adhrai, Ahmad Abdulaziz Al-Owais, Abdelhabib Semlali, and Mazahar Farooqui. 2023. “Antidiabetic, Antioxidant and Cytotoxicity Activities of Ortho- and Para-Substituted Schiff Bases Derived from Metformin Hydrochloride: Validation by Molecular Docking and in Silico ADME Studies.” *Open Chemistry* 21(1):1–15.
- Al-qadsy, Inas, Waseem Sharaf Saeed, Ahmad Abdulaziz Al-owais, Abdelhabib Semlali, Ali Alrabie, Lena Ahmed Saleh Al-faqeeh, Mohammed Alsaeedy, Arwa Al-adhrai, and Mazahar Farooqui. 2023. “Antimicrobial Activity of Novel Ni (II) and Zn (II) Complexes Synthesis , Characterization and Molecular Docking Studies.” (Ii).
- Alabdallah, Nadiyah M. and Md Mahadi Hasan. 2021. “Plant-Based Green Synthesis of Silver Nanoparticles and Its Effective Role in Abiotic Stress Tolerance in Crop Plants.” *Saudi Journal of Biological Sciences* 28(10):5631–39.
- Alahmad, Abdalrahim, Wael A. Al-Zereini, Tahani J. Hijazin, Osama Y. Al-Madanat, Ibrahim Alghoraibi, Omar Al-Qaralleh, Samer Al-Qaraleh, Armin Feldhoff, Johanna Gabriela

- Walter, and Thomas Scheper. 2022. "Green Synthesis of Silver Nanoparticles Using Hypericum Perforatum L. Aqueous Extract with the Evaluation of Its Antibacterial Activity against Clinical and Food Pathogens." *Pharmaceutics* 14(5).
- Alam, Md Zafer, Alimuddin, and Salman A. Khan. 2023. *A Review on Schiff Base as a Versatile Fluorescent Chemo-Sensors Tool for Detection of Cu²⁺ and Fe³⁺ Metal Ion*. Vol. 33. Springer US.
- Alem, Mamaru Bitew, Tegene Desalegn, Tadewos Damena, Enyew Alemayehu Bayle, Moses O. Koobotse, Kennedy J. Ngwira, Japheth O. Ombito, Matshediso Zachariah, and Taye B. Demissie. 2023. "Cytotoxicity and Antibacterial Potentials of Mixed Ligand Cu(II) and Zn(II) Complexes: A Combined Experimental and Computational Study." *ACS Omega* 8(14):13421–34.
- Ali, Mohammad, Bosung Kim, Kevin D. Belfield, David Norman, Mary Brennan, and Gul Shad Ali. 2016. "Green Synthesis and Characterization of Silver Nanoparticles Using Artemisia Absinthium Aqueous Extract - A Comprehensive Study." *Materials Science and Engineering C* 58:359–65.
- Aljeldah, Mohammed Mubarak, Mourad A. M. Aboul-Soud, Mohamed Taha Yassin, and Ashraf Abdel Fattah Mostafa. 2023. "Synergistic Antibacterial Potential of Greenly Synthesized Silver Nanoparticles with Fosfomycin Against Some Nosocomial Bacterial Pathogens." *Infection and Drug Resistance* 16(December 2022):125–42.
- Allahverdiyev, Adil M., Kateryna Volodymyrivna Kon2, Emrah Sefik Abamor1, Malahat Bagirova1, And, and Miriam Rafailovich3. 2011. "Coping with Antibiotic Resistance: Combining Nanoparticles with Antibiotics and Other Antimicrobial Agents." *Expert Rev. Infect. Ther.* 9(11):1035–52.
- Alotaibi, Saad Hmoud and Awad Abdalla Momen. 2019. "Anticancer Drugs' Deoxyribonucleic Acid (DNA) Interactions." Pp. 1–24 in *Biophysical Chemistry - Advance Applications*.
- Altammar, Khadijah A. 2023. "A Review on Nanoparticles: Characteristics, Synthesis, Applications, and Challenges." *Frontiers in Microbiology* 14(April):1–20.
- Ameya Buli, Gemechu. 2015. "Antimicrobial Activity of Lippia Adoensis Var. Koseret Against Human Pathogenic Bacteria and Fungi." *American Journal of Clinical and Experimental Medicine* 3(3):118.

- Anandalakshmi, K., J. Venugobal, and V. Ramasamy. 2016. "Characterization of Silver Nanoparticles by Green Synthesis Method Using Pedalium Murex Leaf Extract and Their Antibacterial Activity." *Applied Nanoscience (Switzerland)* 6(3):399–408.
- Anthon, Elizabeth J., Elizabeth M. Bolitho, a Hannah E. Bridgewater, Oliver W. L. Carter, a Jane M. Donnelly, Cinzia Imberti, Edward C. Lant, Frederik Lermyte, Russell J. Needham, Marta Palau, Peter J. Sadler, Huayun Shi, Fang-Xin Wang, Wen-Ying and Zhang, and Zijin Zhang. 2020. "Metallodrugs Are Unique: Opportunities and Challenges of Discovery and Development." *Royal Society of Chemistry* 11(48):12888–917.
- Araghi, Mahmood, Reza Mannani, Ali Heidarnejad maleki, Adel Hamidi, Samaneh Rostami, Salar Hozhabri Safa, Fatemeh Faramarzi, Sahar Khorasani, Mina Alimohammadi, Safa Tahmasebi, and Reza Akhavan-Sigari. 2023. "Recent Advances in Non-Small Cell Lung Cancer Targeted Therapy; an Update Review." *Cancer Cell International* 23(1):1–26.
- Arnold, Melina, Eileen Morgan, Harriet Rumgay, Allini Mafra, Deependra Singh, Mathieu Laversanne, Jerome Vignat, Julie R. Gralow, Fatima Cardoso, Sabine Siesling, and Isabelle Soerjomataram. 2022. "Current and Future Burden of Breast Cancer : Global Statistics for 2020 and 2040." *The Breast* 66(September):15–23.
- Arya, Geeta, R. Mankamna Kumari, Nidhi Gupta, Ajeet Kumar, Ramesh Chandra, and Surendra Nimesh. 2018. "Green Synthesis of Silver Nanoparticles Using Prosopis Juliflora Bark Extract: Reaction Optimization, Antimicrobial and Catalytic Activities." *Artificial Cells, Nanomedicine and Biotechnology* 46(5):985–93.
- Asif, Muhammad, Riffat Yasmin, Rizwan Asif, Ana Ambreen, and Madiha Mustafa. 2022. "Green Synthesis of Silver Nanoparticles (AgNPs), Structural Characterization , and Their Antibacterial Potential." *Dose-Response: An International Journal* (June):1–11.
- Authors, The. 2024. "1,2* ,." 38(1):147–66.
- Ayala, Hernández, Luis Felipe, Guzmán López, Eduardo Gabriel, and Annia Galano. 2023. "Quinoline Derivatives: Promising Antioxidants with Neuroprotective Potential." *Antioxidants* 12(10):1–17.
- Ayukekbong, James A., Michel Ntemgwa, and Andrew N. Atabe. 2017. "The Threat of Antimicrobial Resistance in Developing Countries : Causes and Control Strategies." 1–8.
- Bairagi, Satyaranjan and Mohammad Reza. 2023. "Review on Green Biomass-Synthesized Metallic Nanoparticles and Composites and Their Photocatalytic Water Purification

- Applications : Progress and Perspectives.” *Chemical Engineering Journal Advances* 14(January):100460.
- Banik, Bhabatosh, Kumar Somyajit, Ganesh Nagaraju, and Akhil R. Chakravarty. 2014. “Cellular Imaging and Mitochondria Targeted.” *Dalton Transactions* 43:13358–69.
- Baquero, Fernando. 2021. “Threats of Antibiotic Resistance : An Obligated Reappraisal.” *International Microbiology* 499–506.
- Becke, Axel D. 1993. “Density-Functional Thermochemistry . III . The Role of Exact Exchange.” *The Journal of Chemical Physics* 98(October 1992):5648–52.
- Behera, Nabakrushna, Tankadhar Behera, Jyotiprabha Rout, and Sasmita Moharana. 2024. “Significant Aspects of Heterocyclic Schiff Bases and Their Metal.” Pp. 1–21 in *INTECH open science*.
- Belachew, G., D. Engeda, and D. Tilku. 2022. “In Vitro Antioxidant Activity of Lippia Adoensis Var. Koseret, Thymus Schimperii Ronniger and Rosmarinus Officinalis Leaf Extracts and Their Effects on Oxidative Stability of Ground Raw Beef Meat during Refrigeration Storage.” *Food Research* 6(5):319–27.
- Bello-vieda, Nestor J., Homero F. Pastrana, Manuel F. Garavito, G. Á. Alba, Adriana M Celis, Alvaro Muñoz-castro, Silvia Restrepo, and John J. Hurtado. 2018. “Antibacterial Activities of Azole Complexes Combined with Silver Nanoparticles.” *Molecules* 23(361):1–17.
- Beyth, Nurit, Yael Hourihaddad, Avi Domb, Wahid Khan, and Ronen Hazan. 2015. “Alternative Antimicrobial Approach : Nano-Antimicrobial Materials.” *Evidence-Based Complementary and Alternative Medicine* 2015:1–16.
- Bheemayya, Lokesh, Ravindra R. Kamble, Arun K. Shettar, Tukaram V. Metre, Barnabas Kodasi, Madivalagouda S. Sannaikar, Sanjeev R. Inamdar, Mussuvir Pasha K. M, and Joy H. Hoskeri. 2023. “Design and Synthesis of Novel Fluorescent 2-(Aryloxy)-3-(4,5-Diaryl)-1H-Imidazol-2-Yl)Quinolines: Solvatochromic, DFT, TD-DFT Studies, COX-1 and COX-2 Inhibition and Antioxidant Properties.” *Journal of Fluorescence* (0123456789).
- Bitew, Mamaru, Tegene Desalegn, Taye B. Demissie, and Anteneh Belayneh. 2021. “Pharmacokinetics and Drug-Likeness of Antidiabetic Flavonoids : Molecular Docking and DFT Study.” *PLoS ONE* 10:1–22.

- Boros, Eszter, Paul J Dyson, and Gilles Gasser. 2020. "Classification of Metal-Based Drugs According to Their Mechanisms of Action." *CHEMPR* 6(1):41–60.
- Boros, Eszter, Paul J. Dyson, and Gilles Gasser. 2020. "Classification of Metal-Based Drugs According to Their Mechanisms of Action." *CHem* 6:41–60.
- Boutayeb, A. 2010. "The Burden of Communicable and Non-Communicable Diseases in Developing Countries." *Handbook of Disease Burdens and Quality of Life Measures* 2010:531–46.
- Brandelli, Adriano. 2015. "Bioactivity of Noble Metal Nanoparticles Decorated with Biopolymers and Their Application in Drug Delivery." *Elsevier B.V.*
- Bruna, Tamara, Francisca Maldonado-Bravo, Paul Jara, and Nelson Caro. 2021. "Silver Nanoparticles and Their Antibacterial Applications." *International Journal of Molecular Sciences* 22(13).
- Burlec, Ana Flavia, Andreia Corciova, Monica Boev, Denisa Batir-Marin, Cornelia Mircea, Oana Cioanca, Gabriela Danila, Marius Danila, Anca Florentina Bucur, and Monica Hancianu. 2023. "Current Overview of Metal Nanoparticles' Synthesis, Characterization, and Biomedical Applications, with a Focus on Silver and Gold Nanoparticles." *Pharmaceuticals* 16(10).
- By, Chaired and J. I. M. O. Neill. 2016. "TACKLING DRUG-RESISTANT INFECTIONS GLOBALLY : FINAL REPORT AND RECOMMENDATIONS THE REVIEW ON." (May).
- Cai, Ling, Yanqiang Huang, Yuanyuan Duan, Qiao Liu, Qilan Xu, Jia Jia, Jianming Wang, and Qian Tong. 2021. "Schiff-Base Silver Nanocomplexes Formation on Natural Biopolymer Coated Mesoporous Silica Contributed to the Improved Curative Effect on Infectious Microbes." 14(8):2735–48.
- Çalhan, Selda Dogan and Mustafa Gundogan. 2020. "Biosynthesis of Silver Nanoparticles Using Onosma Sericeum Willd . and Evaluation of Their Catalytic Properties and Antibacterial and Cytotoxic Activity." *Turkish Journal of Chemistry* 1587–1600.
- ÇALHAN, Selda DOĞAN and Mustafa GÜNDOĞAN. 2020. "Biosynthesis of Silver Nanoparticles Using Onosma Sericeum Willd . and Evaluation of Their Catalytic Properties and Antibacterial and Cytotoxic Activity." *Turkish Journal of Chemistry* 1587–1600.

- Cao, Qing, Ye Tian, Zhiyi Deng, Fangfang Yang, and Erfei Chen. 2024. "Epigenetic Alteration in Colorectal Cancer: Potential Diagnostic and Prognostic Implications." *International Journal of Molecular Sciences* 25(6).
- Caruso, Silvia, Chiara Valenti, Lorella Marinucci, Francesca Di Pasquale, Claudia Truppa, Giulia Di Benedetto, Sara Caruso, and Stefano Pagano. 2024. "Systematic Review of Zinc's Benefits and Biological Effects on Oral Health." *Materilas* 17(800):1–25.
- Chakraborty, Amlan, Jennifer C. Boer, Cordelia Selomulya, and Magdalena Plebanski. 2018. "Amino Acid Functionalized Inorganic Nanoparticles as Cutting-Edge Therapeutic and Diagnostic Agents." *Bioconjugate Chemistry* 29(3):657–71.
- Chandra, Abhishek and Man Singh. 2018. "Biosynthesis of Amino Acid Functionalized Silver Nanoparticles for Potential Catalytic and Oxygen Sensing Applications." *Inorganic Chemistry Frontiers* 5(1):233–57.
- Chandra, Sulekh and Umendra Kumar. 2005. "Spectral and Magnetic Studies on Manganese(II), Cobalt(II) and Nickel(II) Complexes with Schiff Bases." *Spectrochimica Acta - Part A: Molecular and Biomolecular Spectroscopy* 61(1–2):219–24.
- Chandrakala, V., Valmiki Aruna, and Gangadhara Angajala. 2022. "Review on Metal Nanoparticles as Nanocarriers: Current Challenges and Perspectives in Drug Delivery Systems." *Emergent Materials* 5(6):1593–1615.
- Chaudhary, Priya, Pracheta Janmeda, Anca Oana Docea, Balakyz Yeskaliyeva, Ahmad Faizal, Abdull Razis, Babagana Modu, Daniela Calina, and Javad Shari. 2023. "Oxidative Stress , Free Radicals and Antioxidants : Potential Crosstalk in the Pathophysiology of Human Diseases." *Frontiers in Chemistry* (May):1–24.
- Chebout, Oussama, Chahrazed Trifa, Sofiane Bouacida, and Mhamed Boudraa. 2022. "Two New Copper (II) Complexes with Sulfanilamide as Ligand : Synthesis , Structural , Thermal Analysis , Electrochemical Studies and Antibacterial Activity." *Journal of Molecular Structure* 1248:131446.
- Chen, C. H., P. A. Campbell, and L. S. Newman. 1990. "MTT Colorimetric Assay Detects Mitogen Responses of Spleen but Not Blood Lymphocytes." *Int Arch Allergy Appl Immunol* 93(2–3):249–55.
- Chen, Yu and Longqin Hu. 2008. "Design of Anticancer Prodrugs for Reductive Activation." *Medicinal Research Reviews* 29(1):29–64.

- Chikkanayakanahalli Paramesh, Chitrabanu, Guddappa Halligudra, Vinay Gangaraju, Jagadeesh Babu Sriramoju, Mahesh Shastri, Harsha Kachigere B., Preetham Habbanakuppe D., Dinesh Rangappa, Rangappa Kanchugarakoppal Subbegowda, and Prasanna Doddakunche Shivaramu. 2021. "Silver Nanoparticles Synthesized Using Saponin Extract of Simarouba Glauca Oil Seed Meal as Effective, Recoverable and Reusable Catalyst for Reduction of Organic Dyes." *Results in Surfaces and Interfaces* 3(January):100005.
- Chkirate, Karim, Khalid Karrouchi, Necmi Dege, Nada Kheira Sebbar, Abdelaziz Ejjoumany, Smaail Radi, N. N. Adarsh, Ahmed Talbaoui, Marilena Ferbinteanu, El Mokhtar Essassi, and Yann Garcia. 2020. "Co(Ii) and Zn(Ii) Pyrazolyl-Benzimidazole Complexes with Remarkable Antibacterial Activity." *New Journal of Chemistry* 44(6):2210–21.
- ChunYan, Zhang, Yu RuJian, Wang LiQiang, Huang HaiYan, Wang JinTao, Liao XiangWen, Duan XueMin, and Xiong YanShi. 2022. "Design, Synthesis, and Evaluation of Aryl-Thioether Ruthenium Polypyridine Complexes: A Multi-Target Antimicrobial Agents against Gram-Positive Bacteria." *European Journal of Medicinal Chemistry* 240:114562.
- Ciccotti, G., C. Dellago, M. Ferrario, and E. R. Hern. 2022. "Molecular Simulations : Past , Present , and Future (a Topical Issue in EPJB)." *THE EUROPEAN PHYSICAL JOURNAL B* 95(3):1–12.
- Claudel, Mickaël, Justine V Schwarte, and Katharina M. Fromm. 2020. "New Antimicrobial Strategies Based on Metal Complexes." *Chemistry* 2:849–99.
- Cohen, Seth M. 2007. "New Approaches for Medicinal Applications of Bioinorganic Chemistry." *Current Opinion in Chemical Biology* 11(II):115–20.
- Coque, Teresa M., Rafael Cant, Ana Elena P, Miguel D. Fern, and Fernando Baquero. 2023. "Antimicrobial Resistance in the Global Health Network : Known Unknowns and Challenges for Efficient Responses in the 21st Century."
- Corsi, Ilaria, Martin Federico Desimone, and Jimena Cazenave. 2022. "Building the Bridge From Aquatic Nanotoxicology to Safety by Design Silver Nanoparticles." *Ontiers in Bioengineering and Biotechnology* 10(March):1–28.
- Dakal, Tikam Chand, Anu Kumar, Rita S. Majumdar, and Vinod Yadav. 2016. "Mechanistic Basis of Antimicrobial Actions of Silver Nanoparticles." *Frontiers in Microbiology*

7(NOV):1–17.

- Damena, Tadewos, Mamaru Bitew Alem, Digafie Zeleke, Tegene Desalegn, Rajalakshmanan Eswaramoorthy, and Taye B. Demissie. 2022a. “Novel Zinc(II) and Copper(II) Complexes of 2-((2-Hydroxyethyl)Amino)Quinoline-3-Carbaldehyde for Antibacterial and Antioxidant Activities: A Combined Experimental, DFT, and Docking Studies.” *ACS Omega* 7(30):26336–52.
- Damena, Tadewos, Mamaru Bitew Alem, Digafie Zeleke, Tegene Desalegn, Rajalakshmanan Eswaramoorthy, and Taye B. Demissie. 2022b. “Synthesis, Characterization, and Biological Activities of Zinc(II), Copper(II) and Nickel(II) Complexes of an Aminoquinoline Derivative.” *Frontiers in Chemistry* 10(November):1–21.
- Danielsen, Anders Skyrud, Léa Franconeri, Samantha Page, Anders Eivind Myhre, Ragnhild Agathe Ternes, Oliver Kacelnik, and Jørgen Vildershøj Bjørnholt. 2023. “Clinical Outcomes of Antimicrobial Resistance in Cancer Patients: A Systematic Review of Multivariable Models.” *BMC Infectious Diseases* 23(1):1–16.
- Dasari, S., & Bernard T., P. (2014). Cisplatin in cancer therapy: Molecular mechanisms of action. *European Journal of Pharmacology*, 740, 364–378.
- Dhabale, Raj Hasamukh, Shalini Shah, Neha Tiwari, and Pragnesh Patani. 2022. “Review Of Semicarbazone , Thiosemicarbazone , And Their Transition Metal Complexes , And Their Biological Activities.” 13(5):2416–24.
- Dhanda, Geetika, Yash Acharya, and Jayanta Haldar. 2023. “Antibiotic Adjuvants: A Versatile Approach to Combat Antibiotic Resistance.” *ACS Omega* 8(12):10757–83.
- Dharmaraj, Divya, Madhuri Krishnamoorthy, Kumar Rajendran, Kannan Karuppiah, Jeyameenakshi Annamalai, Karthick Rajan Durairaj, Prakash Santhiyagu, and Kannapiran Ethiraj. 2021. *Antibacterial and Cytotoxicity Activities of Biosynthesized Silver Oxide (Ag₂O) Nanoparticles Using Bacillus Paramycoides*. Vol. 61. Elsevier B.V.
- Dhingra, Sameer, Nor Azlina A. Rahman, Ed Peile, and Motiur Rahman. 2020. “Microbial Resistance Movements: An Overview of Global Public Health Threats Posed by Antimicrobial Resistance , and How Best to Counter.” 8(November):1–22.
- Dine, Ilili, Endale Mulugeta, Yadessa Melaku, and Melis Belete. 2023. “Recent Advances in the Synthesis of Pharmaceutically Active 4-Quinolone and Its Analogues: A Review.” *RSC Advances* 13(13):8657–82.

- Dinku, Dadi, Taye B. Demissie, Isaac N. Beas, Rajalakshmanan Eswaramoorthy, Bayan Abdi, and Tegene Desalegn. 2024. "Antimicrobial Activities and Docking Studies of New Schiff Base Ligand and Its Cu (II), Zn (II) and Ni (II) Complexes : Synthesis and Characterization." *Inorganic Chemistry Communications* 160.
- Egodawaththa, Nishal M., Amy L. Knight, Jingxuan Ma, D. Andrew Knight, Eric Guisbert, and Nasri Nesnas. 2022. "Synthesis and Characterization of Ligand-Stabilized Silver Nanoparticles and Comparative Antibacterial Activity against E. Coli." *International Journal of Molecular Sciences* 23(23).
- Eichhorn, Thomas, Dušan Dimić, and Zoran Marković. 2023. "Synthesis , Spectroscopic Characterization and DFT Analysis of Dichlorido (η^6 - p -Cymene) Ruthenium (II) Complexes with Isonicotinate-Polyethylene Glycol Ester Ligands." *Journal of the Serbian Chemical Society* 88(December):1335–54.
- Ejidike, Ikechukwu P. 2018. "Cu(II) Complexes of 4-[(1E)-N-{2-[(Z)-Benzylidene-Amino]Ethyl} Ethanimidoyl]Benzene-1,3-Diol Schiff Base: Synthesis, Spectroscopic, in-Vitro Antioxidant, Antifungal and Antibacterial Studies." *Molecules* 23(7):1–18.
- El-Lateef, Hany M. Ab., Tarek El-Dabea, Mai M. Khalaf, and Ahmed M. Abu-Dief. 2023. "Recent Overview of Potent Antioxidant Activity of Coordination Compounds." *Antioxidants* 12(2).
- Elebiju, Oluwadunni F., Olayinka O. Ajani, Gbolahan O. Oduselu, and Temitope A. Oggunpebi. 2023. "Recent Advances in Functionalized Quinoline Scaffolds and Hybrids — Exceptional Pharmacophore in Therapeutic Medicine." *Frontiers in Chemistry* (January):1–18.
- Eleonora, Mayra, Beltrán Pineda, Luz Marina, Lizarazo Forero, Cesar Augusto, and Sierra Avila. 2022a. "Antibacterial Activity of Biosynthesized Silver Nanoparticles (AgNps) against Pectobacterium Carotovorum." *Brazilian Journal of Microbiology* 1175–86.
- Eleonora, Mayra, Beltrán Pineda, Luz Marina, Lizarazo Forero, Cesar Augusto, and Sierra Avila. 2022b. "Antibacterial Activity of Biosynthesized Silver Nanoparticles (AgNps) against Pectobacterium Carotovorum." *Brazilian Journal of Microbiology* 1175–86.
- Ershov, Vadim, Natalia Tarasova, and Boris Ershov. 2021. "Evolution of Electronic State and Properties of Silver Nanoparticles during Their Formation in Aqueous Solution." *International Journal of Molecular Sciences* 22(19).

- Essawy, Ehab, Mohamed S. Abdelfattah, Mansour El-Matbouli, and Mona Saleh. 2021. "Synergistic Effect of Biosynthesized Silver Nanoparticles and Natural Phenolic Compounds against Drug-Resistant Fish Pathogens and Their Cytotoxicity: An in Vitro Study." *Marine Drugs* 19(1).
- Evans, Andris and Kevin A. Kavanagh. 2021. "Evaluation of Metal-Based Antimicrobial Compounds for the Treatment of Bacterial Pathogens." *Journal of Medical Microbiology* 70(5):1–18.
- Excellence, I. C. Verma and Young Pediatricians. 2022. "Antimicrobial Resistance: The 'Other' Pandemic! What Are the Reasons for Antimicrobial." *Indian Journal of Pediatrics* 89(June):600–606.
- Fayyadh, Ali Abdullah and Muneer H. Jaduaa Alzubaidy. 2021. "Green-Synthesis of Ag₂O Nanoparticles for Antimicrobial Assays." *Journal of the Mechanical Behavior of Materials* 30(1):228–36.
- Ferlay, Jacques, Murielle Colombet, Isabelle Soerjomataram, Donald M. Parkin, Marion Piñeros, Ariana Znaor, and Freddie Bray. 2024. "Cancer Statistics for the Year 2020: An Overview." *International Journal of Cancer* 149(4):778–89.
- Fern, Sandra, Isabel Velo-helena, Miguel Mart, Ramiro Barcia, Ò. Palacios, Merc Capdevila, Ana M. Gonz, and Rosa Pedrido. 2023. "Exploring the Biological Properties of Zn (II) Bis Thiosemicarbazone Helicates NCI-H460 The Ligands Were." (li).
- Fonseca-l, Daniela, Johan D. Lozano, Mario A. Mac, Á. Muñoz-castro, Desmond Macleod-carey, Edgar Nagles, and John Hurtado. 2024. "Biological Activity of Complexes Involving Nitro-Containing Ligands and Crystallographic-Theoretical Description of 3 , 5-DNB Complexes."
- Fratoddi, Ilaria, Iole Venditti, Chiara Battocchio, Laura Carlini, Simone Amatori, and Carlo Santini. 2019. "Highly Hydrophilic Gold Nanoparticles as Carrier for Anticancer Copper (I) Complexes: Loading and Release Studies for Biomedical Applications." *Nanomaterials* 9(I):1–13.
- Frisch, M. J., G. W. Trucks, H. B. Schlegel, G. E. Scuseria, M. A. Robb, J. R. Cheeseman, G. Scalmani, V. Barone, G. A. Petersson, H. Nakatsuji, X. Li, M. Caricato, A. V. Marenich, J. Bloino, B. G. Janesko, Gomperts, R.M., and E.U.A. 2016. "Gaussian, Inc., Wallingford CT." *Gaussian 16, Revision B.01*.

- Gamberi, Tania and Muhammad Hanif. 2022. "Metal-Based Complexes in Cancer Treatment." *Biomedicines* 10(10):6–8.
- Geisler, Heiko, Julia Westermayr, Klaudia Cseh, Dominik Wensch, Valentin Fuchs, Sophia Harringer, Sarah Plutzar, Natalie Gajic, Michaela Hejl, Michael A. Jakupiec, Philipp Marquetand, and Wolfgang Kandioller. 2021. "Tridentate 3-Substituted Naphthoquinone Ruthenium Arene Complexes: Synthesis, Characterization, Aqueous Behavior, and Theoretical and Biological Studies." *Inorganic Chemistry* 60(13):9805–19.
- Gemechu, Shumi, Tegene Desalegn, Taye B. Demissie, Venkatesha Perumal Ramachandran, and Rajalakshmanan Eswaramoorthy. 2022. "Metal Complexes in Target-Specific Anticancer Therapy : Recent Trends and Challenges." *Journal of Chemistry* 2022:1–19.
- Giri, Alok Kumar, Biswajit Jena, Bhagyashree Biswal, Arun Kumar Pradhan, Manoranjan Arakha, Saumyapraava Acharya, and Laxmikanta Acharya. 2022. "Green Synthesis and Characterization of Silver Nanoparticles Using Eugenia Roxburghii DC. Extract and Activity against Biofilm-Producing Bacteria." *Scientific Reports* 12(1):1–9.
- Gonzalez-Ballesteros, Mauricio M., Carmen Mejia, and Lena Ruiz-Azuara¹. 2022. "Metallodrugs : An Approach against Invasion and Metastasis in Cancer Treatment." *FEBS Open Bio* 12:880–99.
- González-Larraz, Pamela G., Tessy M. López-Goerne, Francisco J. Padilla-Godínez, Marco A. González-López, Aida Hamdan-Partida, and Esteban Gómez. 2020. "IC₅₀ Evaluation of Platinum Nanocatalysts for Cancer Treatment in Fibroblast, HeLa, and DU-145 Cell Lines." *ACS Omega* 5(39):25381–89.
- Gopichand, Kamble, Varukolu Mahipal, N. Nageswara Rao, Abdul Majeed Ganai, and P. Venkateswar Rao. 2023. "Results in Chemistry Co (II), Ni (II), Cu (II), and Zn (II) Complexes with Benzothiazole Schiff Base Ligand : Preparation , Spectral Characterization , DNA Binding , and In Vitro Cytotoxic Activities." *Results in Chemistry* 5(December 2022):100868.
- Goto, Masuo, Jun-cai Li, Guan-zhou Yang, Kuo-hsiung Lee, North Carolina, Chapel Hill, and North Carolina. 2019. "Biologically Active Quinoline and Quinazoline Alkaloids Part I." *Medical Research Review* 38(3):775–828.
- Grimme, Stefan. 2002. "An Improved Method for Density Functional Calculations of the Frequency-Dependent Optical Rotation." *Chemical Physics Letters* 361(July):321–28.

- Guan, Defeng, Lihui Zhao, Xin Shi, Xiaoling Ma, and Zhou Chen. 2023. "Copper in Cancer : From Pathogenesis to Therapy." *Biomedicine & Pharmacotherapy* 163(April):1–17.
- Guerrini, Luca, Ramon A. Alvarez-Puebla, and Nicolas Pazos-Perez. 2018. "Surface Modifications of Nanoparticles for Stability in Biological Fluids." *Materials* 11(7):1–28.
- Gul, Rahman, Syed Umer Jan, Syed Faridullah, Samiullah Sherani, and Nusrat Jahan. 2017. "Preliminary Phytochemical Screening, Quantitative Analysis of Alkaloids, and Antioxidant Activity of Crude Plant Extracts from Ephedra Intermedia Indigenous to Balochistan." *Scientific World Journal* 2017(Figure 1).
- Gunawan, Ramdhan, Asep Bayu, and Dani Nandiyanto. 2021. "How to Read and Interpret ¹H-NMR and ¹³C-NMR Spectrums." *Indonesian Journal of Science & Technology* 6(2):267–98.
- Gupta, Priyamvada, Nilesh Rai, Ashish Verma, Dimple Saikia, Surya Pratap Singh, Rajiv Kumar, Santosh Kumar Singh, Deepak Kumar, and Vibhav Gautam. 2022. "Green-Based Approach to Synthesize Silver Nanoparticles Using the Fungal Endophyte *Penicillium Oxalicum* and Their Antimicrobial, Antioxidant, and in Vitro Anticancer Potential." *ACS Omega* 7(50):46653–73.
- Hajfathalian, Maryam, Katherine J. Mossburg, Allan Radaic, Katherine E. Woo, Pallavi Jonnalagadda, Yvonne Kapila, and Paul L. Bollyky. 2024. "A Review of Recent Advances in the Use of Complex Metal Nanostructures for Biomedical Applications from Diagnosis to Treatment." *WIREs Nanomedicine and Nanobiotechnology* (March):1–55.
- Hajipour, Mohammad J., Katharina M. Fromm, Ali Akbar Ashkarran, Dorleta Jimenez de Aberasturi, Idoia Ruiz de Larramendi, Teofilo Rojo, Vahid Serpooshan, Wolfgang J. Parak, and Morteza Mahmoudi. 2012. "Antibacterial Properties of Nanoparticles." *Trends in Biotechnology* 30(10):499–511.
- Hamama, Wafaa S., Mona E. Ibrahim, Ayaa A. Gooda, and Hanafi H. Zoorob. 2018. "Recent Advances in the Chemistry of 2-Chloroquinoline-3-Carbaldehyde and Related Analogs." *RSC Advances* 8(16):8484–8515.
- Hambleton, Ian R., Roberta Caixeta, Selvi M. Jeyaseelan, Silvana Luciani, and Anselm J. M. Hennis. 2023. "The Rising Burden of Non-Communicable Diseases in the Americas and the Impact of Population Aging: A Secondary Analysis of Available Data." *The Lancet Regional Health - Americas* 21:100483.

- Hassan, Ali M., Ahmed O. Said, Bassem H. Heakal, Ahmed Younis, Wael M. Aboulthana, and Mohamed F. Mady. 2022. "Green Synthesis, Characterization, Antimicrobial and Anticancer Screening of New Metal Complexes Incorporating Schiff Base." *ACS Omega* 7(36):32418–31.
- Hawila, Saly, Florian Massuyeau, Romain Gautier, Alexandra Fateeva, Sébastien Lebegue, Won June Kim, Gilles Ledoux, Adel Mesbah, and Aude Demessence. 2023. "Tuning the 1D–2D Dimensionality upon Ligand Exchange in Silver Thiolate Coordination Polymers with Photoemission Switching." *Journal Materials Chemistry B* 11:3979–84.
- Hay, P, Jeffrey and and Willard R. Wadt. 1985. "Ab Initio Effective Core Potentials for Molecular Calculations . Potentials for the Transition Metal Atoms Sc to Hg Ab Initio Effective Core Potentials for Molecular Calculations . Potentials for the Transition Metal Atoms Sc to Hg." *The Journal of Chemical Physics* 270(82):270–83.
- Hernández, Wilfredo, Fernando Carrasco, Abraham Vaisberg, Evgenia Spodine, Jorge Manzur, Maik Icker, Harald Krautscheid, and Lothar Beyer. 2020. "Synthesis, Spectroscopic Characterization, Structural Studies, and in Vitro Antitumor Activities of Pyridine-3-Carbaldehyde Thiosemicarbazone Derivatives." *Journal of Chemistry* 2020:1–12.
- Hieu, H. N., D. T. H. Trang, V. T. T. Hien, N. V. Nghia, N. T. Lam, and T. M. D. Nguyen. 2022. "Microorganism-Mediated Green Synthesis of Silver Nanoparticles Using *Aspergillus Niger* and *Bacillus Megaterium*." *Digest Journal of Nanomaterials and Biostructures* 17(1):359–67.
- Holden, Lorcan, Christopher S. Burke, David Cullinane, and Tia E. Keyes. 2021. "Strategies to Promote Permeation and Vectorization, and Reduce Cytotoxicity of Metal Complex Luminophores for Bioimaging and Intracellular Sensing." *RSC Chemical Biology* 2(4):1021–49.
- Hong, Yuhang, Alessandra Boiti, Daniela Vallone, and Nicholas S. Foulkes. 2024. "Reactive Oxygen Species Signaling and Oxidative Stress: Transcriptional Regulation and Evolution." *Antioxidants* 13(312):1–24.
- Htwe, Y. Z. N., W. S. Chow, Y. Suda, and M. Mariatti. 2019. "Effect of Silver Nitrate Concentration on the Production of Silver Nanoparticles by Green Method." *Materials Today: Proceedings* 17:568–73.

- Hu, Chun Chih, Gong Her Wu, Sheng Feng Lai, Muniesh Muthaiyan Shanmugam, Y. Hwu, Oliver I. Wagner, and Ta Jen Yen. 2018. "Toxic Effects of Size-Tunable Gold Nanoparticles on *Caenorhabditis Elegans* Development and Gene Regulation." *Scientific Reports* 8(1):1–10.
- Hudzicki, Jan. 2009. "Kirby-Bauer Disk Diffusion Susceptibility Test Protocol Author Information." *American Society For Microbiology* (December 2009):1–23.
- Humphries¹, Romney M., April N. Abbott, and Janet A. Hindler. 2019. "Understanding and Addressing CLSI Breakpoint Revisions - A Primer for Clinical Laboratories." *Journal of Clinical Microbiology* (April):1–36.
- Hussain, Zainab, Emad Yousif, Ahmed Ahmed, and Ali Altaie. 2014. "Synthesis and Characterization of Schiff ' s Bases of Sulfamethoxazole." *Organic and Medicinal Chemistry Letters* 4(1):1–4.
- Ibrahim, Ahmed B. M., Micheal K. Farh, and Peter Mayer. 2018. "Copper Complexes of New Thiosemicarbazone Ligands: Synthesis, Structural Studies and Antimicrobial Activity." *Inorganic Chemistry Communications* #pagerange#.
- Ibrahim, Syed, Nagaraja Naik, Chandan Shivamallu, H. L. Raghavendra, Ali A. Shati, Mohammad Y. Alfaifi, Serag Eldin, I. Elbehairi, Raghavendra G. Amachawadi, and Shiva Prasad. 2024. "Inorganica Chimica Acta Synthesis , Structure , and in Vitro Biological Studies of Benzothiazole Based Schiff Base Ligand and Its Binary and Ternary Co (III) and Ni (II) Complexes." *Inorganica Chimica Acta* 559(August 2023):121792.
- Ismael, Mohammed, Mostafa K. Rabia, and Aly Abdou. 2020. "Design and Synthesis of Three Fe(III) Mixed-Ligand Complexes: Exploration of Their Biological and Phenoxazinone Synthase-Like Activities." *Inorganica Chimica Acta* (Iii):119443.
- Jagessar RC. 2023. "Metal Ions, Ligands and Complexes in Medicinal Chemistry." *Crimson Publishers* 4(1):170–75.
- Jamkhande, Prasad Govindrao, Namrata W. Ghule, Abdul Haque Bamer, and Mohan G. Kalaskar. 2019. "Metal Nanoparticles Synthesis: An Overview on Methods of Preparation, Advantages and Disadvantages, and Applications." *Journal of Drug Delivery Science and Technology* 53:101174.
- Jayaseelan, Poomalai, Selladurai Prasad, Subramanian Vedanayaki, and Rangappan Rajavel.

2016. "Synthesis, Characterization, Anti-Microbial, DNA Binding and Cleavage Studies of Schiff Base Metal Complexes." *Arabian Journal of Chemistry* 9:S668–77.
- Jazzazi, Taghreed M. A., Tareq M. A. Al-Shboul, Mohammad El-khateeb, Hayato Moriyama, Yukihiro Yoshida, and Hiroshi Kitagawa. 2024. "Synthesis, Characterization and Crystallographic Determination of Symmetrical Schiff Bases and Their Zn(II) Metal Complexes." *Transition Metal Chemistry* (Ii).
- Jiang, X. C., W. M. Chen, C. Y. Chen, S. X. Xiong, and A. B. Yu. 2011. "Role of Temperature in the Growth of Silver Nanoparticles Through a Synergetic Reduction Approach." *Nanoscale Research Letters* 6(32):1–9.
- Johnson, Martha S., Preeyaporn Songkiatisak, Pavan Kumar Cherukuri, and Xiao-hong Nancy Xu. 2022. "Toxic Effects of Silver Ions on Early Developing Zebrafish Embryos Distinguished from Silver Nanoparticles." *ACS Omega* 7:40446–55.
- Jomova, Klaudia, Renata Raptova, Suliman Y. Alomar, Saleh H. Alwasel, Eugenie Nepovimova, Kamil Kuca, and Marian Valko. 2023. *Reactive Oxygen Species, Toxicity, Oxidative Stress, and Antioxidants: Chronic Diseases and Aging*. Vol. 97. Springer Berlin Heidelberg.
- Joseph, Tomy Muringayil, Debarshi Kar Mahapatra, Amin Esmaeili, Łukasz Piszczyk, Mohamed S. Hasanin, Mashhoor Kattali, Józef Haponiuk, and Sabu Thomas. 2023. "Nanoparticles: Taking a Unique Position in Medicine." *Nanomaterials* 13(3).
- Joy, Jicksy, Mithun Sanadi Gurumurthy, Riya Thomas, and Manoj Balachandran. 2021. "Biosynthesized AG Nanoparticles: A Promising Pathway for Bandgap Tailoring." *Biointerface Research in Applied Chemistry* 11(2):8875–83.
- Jurca, Tünde, Eleonora Marian, Laura Grațîela, Mariana Eugenia Mureșan, Tünde Jurca, Eleonora Marian, and Laura Grațîela Vicaș. 2017. "Metal Complexes of Pharmaceutical Substances." Pp. p123-142 in *Spectroscopic Analyses - Developments and Applications chemistry*.
- Karges, Johannes, Ryjul W. Stokes, and Seth M. Cohen. 2022. "Metal Complexes for Therapeutic Applications." *Trends in Chemistry* 3(7):523–34.
- Karthik, C., S. Suresh, G. Sneha Mirulalini, and S. Kavitha. 2020. "A FTIR Approach of Green Synthesized Silver Nanoparticles by Ocimum Sanctum and Ocimum Gratissimum on Mung Bean Seeds." *Inorganic and Nano-Metal Chemistry* 50(8):606–12.

- Karthika J. Kadassery, a Matthew R. Crawley, a Samantha N. MacMillan, b David C. Lacy. 2020. "A Hemilabile Manganese (I) -Phenol Complex and Its Adjust Margins ARTICLE A Hemilabile Manganese (I) -Phenol Complex and Its Coordination Induced O – H Bond Weakening." *Dalton Transactions* 00(I):1–10.
- Kaur, Ramandeep and Kapil Kumar. 2021. "Synthetic and Medicinal Perspective of Quinolines as Antiviral Agents." *European Journal of Medicinal Chemistry Journal* 215(January):1–40.
- Kayed, Kamal and Ghaytha Mansour. 2022. "The Antimicrobial Activity of Silver Nanoparticles in Ag/Ag₂ o Composites Synthesized by Oxygen Plasma Treatment of Silver Thin Films." *Current Applied Science and Technology* 22(2):1439–49.
- Kerr, Sheena C., Molly M. Morgan, Amani A. Gillette, Megan K. Livingston, Karina M. Lugo-Cintrón, Peter F. Favreau, Logan Florek, Brian P. Johnson, Joshua M. Lang, Melissa C. Skala, and David J. Beebe. 2020. "A Bioengineered Organotypic Prostate Model for the Study of Tumor Microenvironment-Induced Immune Cell Activation." *Integrative Biology* 12(10):250–62.
- Khakhlary, Prithviraj and Jubaraj B. Baruah. 2015. "Studies on Cluster, Salt and Molecular Complex of Zinc-Quinolate." *Journal of Chemical Sciences* 127(2):215–23.
- Khalaf, Mai M., Hany M. Abd El-lateef, Mohamed Gouda, and Fatma N. Sayed. 2022. "Design , Structural Inspection and Bio-Medicinal Applications of Some Novel Imine Metal Complexes Based on Acetylferrocene." *Materials* 15(4842):1–19.
- Khaldoun, Khadija, Sumera Khizar, Salima Saidi-Besbes, Nadia Zine, Abdelhamid Errachid, and Abdelhamid Elaissari. 2024. "Synthesis of Silver Nanoparticles as an Antimicrobial Mediator." *Journal of Umm Al-Qura University for Applied Sciences* (0123456789).
- Khalil, Eman A. M., Walaa H. Mahmoud, Maher M. I. El Desssouky, and Gehad G. Mohamed. 2021. "Synthesis, Spectral, Thermal and Biological Studies of Some Transition and Inner Transition Schiff Base Metal Complexes." *Egyptian Journal of Chemistry* 64(7):3555–71.
- Khalil, Saied M. E., Magdy Shebl, and Faizah S. Al-gohani. 2014. "Zinc (II) Thiosemicarbazone Complex As a Ligand Towards Some Transition Metal Ions : Synthesis , Spectroscopic and Antimicrobial Studies." (September 2010).
- Khoza, Phindile and Tebello Nyokong. 2015. "Journal of Molecular Catalysis A : Chemical

- Photocatalytic Behaviour of Zinc Tetraamino Phthalocyanine-Silver Nanoparticles Immobilized on Chitosan Beads.” *Journal of Molecular Catalysis. A, Chemical* 399:25–32.
- Kiymaci, Merve Eylul, Hakan Erdoğan, and Merve Bacanlı. 2022. “Antioxidative, Cytotoxic, and Antibacterial Properties of Self-Assembled Glycine-Histidine-Based Dipeptides with or without Silver Nanoparticles in Bio-Inspired Film.” *Arhiv Za Higijenu Rada i Toksikologiju* 73(2):169–77.
- Klębowski, Bartosz, Joanna Depciuch, Magdalena Parlińska-Wojtan, and Jarek Baran. 2018. “Applications of Noble Metal-Based Nanoparticles in Medicine.” *International Journal of Molecular Sciences* 19(12).
- Kong, Fen-Ying, Jin-Wei Zhang, Rong-Fang Li, Zhong-Xia Wang, Wen-Juan Wang, and Wei Wang. 2017. “Unique Roles of Gold Nanoparticles in Drug Delivery , Targeting and Imaging Applications.” *Molecules* 22(1445):1–13.
- Kostova, Irena. 2023. “The Role of Complexes of Biogenic Metals in Living Organisms.” *Inorganics Review* 11(56):1–30.
- Kotrange, Harshada, Agnieszka Najda, Aarti Bains, Robert Gruszecki, Prince Chawla, and Mansuri M. Tosif. 2021. “Metal and Metal Oxide Nanoparticle as a Novel Antibiotic Carrier for the Direct Delivery of Antibiotics.” *International Journal of Molecular Sciences* 22(17).
- Krishnan, R., J. S. Binkley, R. Seeger, and J. A. Pople. 1980. “Self-Consistent Molecular Orbital Methods. XX. A Basis Set for Correlated Wave Functions.” *The Journal of Chemical Physics* 72(1):650–54.
- Kulkarni, Deepak, Rushikesh Sherkar, Chaitali Shirsathe, Rushikesh Sonwane, Nikita Varpe, Santosh Shelke, Mahesh P. More, Sagar R. Pardeshi, Gargee Dhaneshwar, Vijayabhaskarreddy Junnuthula, and Sathish Dyawanapelly. 2023. “Biofabrication of Nanoparticles: Sources, Synthesis, and Biomedical Applications.” *Frontiers in Bioengineering and Biotechnology* 11(May):1–26.
- Kumah, Elizabeth Adjoa, Raoul Djou Fopa, Saeed Harati, Paul Boadu, Fatemeh Vida Zohoori, and Tannaz Pak. 2023a. “Human and Environmental Impacts of Nanoparticles: A Scoping Review of the Current Literature.” *BMC Public Health* 23(1).
- Kumah, Elizabeth Adjoa, Raoul Djou Fopa, Saeed Harati, Paul Boadu, Fatemeh Vida Zohoori,

- and Tannaz Pak. 2023b. "Human and Environmental Impacts of Nanoparticles: A Scoping Review of the Current Literature." *BMC Public Health* 23(1).
- Kumar, Munish, Kavita Bansal, Vijay S. Gondil, Samrita Sharma, D. V. S. Jain, Sanjay Chhibber, Rohit K. Sharma, and Nishima Wangoo. 2018. "Synthesis, Characterization, Mechanistic Studies and Antimicrobial Efficacy of Biomolecule Capped and PH Modulated Silver Nanoparticles." *Journal of Molecular Liquids* 249:1145–50.
- Kumar, Nishitha R., Tejashree A. Balraj, Swetha N. Kempegowda, and Akila Prashant. 2024. "Multidrug-Resistant Sepsis: A Critical Healthcare Challenge." *Antibiotics* 13(1):1–38.
- Kumar, Priti, Arvindhan Nagarajan, and Pradeep D. Uchil. 2018. "Analysis of Cell Viability by the MTT Assay." *Cold Spring Harbor Protocols* 2018(6):469–71.
- Kumar, Singh Virendra, Sharma Pramod Kumar, Dudhe Rupesh, and Kumar Nitin. 2011. "Synthesis, Characterization and Biological Studies of Co(II), Ni(II), Cu(II) and Zn(II) Complexes of Schiff Bases Derived from 3-Formyl-2-Mercaptoquinolines." *Journal of Chemical and Pharmaceutical Research* 3(1):675–84.
- Laouini, Salah Eddine, Abderrhmane Bouafia, Alexander V. Soldatov, Hamed Algarni, Mohammed Laid Tedjani, Gomaa A. M. Ali, and Ahmed Barhoum. 2021. "Green Synthesized of Ag/Ag₂O Nanoparticles Using Aqueous Leaves Extracts of Phoenix Dactylifera l. And Their Azo Dye Photodegradation." *Membranes* 11(7).
- Lee, Chengteh, Chapel Hill, and North Carolina. 1988. "Development of the Colic-Salvetti Correlation-Energy Formula into a Functional of the Electron Density." *Physical Review B* 37(2):785–89.
- Li, Guodong, Hao Liu, Ruibing Feng, Tian Shu Kang, Wanhe Wang, Chung Nga Ko, Chun Yuen Wong, Min Ye, Dik Lung Ma, Jian Bo Wan, and Chung Hang Leung. 2021. "A Bioactive Ligand-Conjugated Iridium(III) Metal-Based Complex as a Keap1–Nrf2 Protein-Protein Interaction Inhibitor against Acetaminophen-Induced Acute Liver Injury." *Redox Biology* 48:102129.
- Liaqat, Nida, Nazish Jahan, Khalil-ur-Rahman, Tauseef Anwar, and Huma Qureshi. 2022. "Green Synthesized Silver Nanoparticles: Optimization, Characterization, Antimicrobial Activity, and Cytotoxicity Study by Hemolysis Assay." *Frontiers in Chemistry* 10(August):1–13.
- Liu, Jiatong, Xiaoyue Han, Tingyue Zhang, Keyue Tian, Zhaoping Li, and Feng Luo. 2023.

- “Reactive Oxygen Species (ROS) Scavenging Biomaterials for Anti - Inflammatory Diseases : From Mechanism to Therapy.” *Journal of Hematology & Oncology* 16(116):1–34.
- Liu, Pei, Wen-Hao Zhao, Yang-Ping Song, Qiang Wang, Yi-Fan Kan, Shi-Yan Wang, Ji-Lin Xia, Muhammad Bilal, Xiao-Yan Zhu, Zhong-Xing Wang, Min Sun, Shuai He, and Xiang-Qian Li. 2022. “Characterization, Antimicrobial, and Antioxidant Potentialities of First-Time Isolated Silver Nanoparticles Synthesizing Protein Secreted by *Lysinibacillus Sphaericus*.” *Process Biochemistry* 122(1):230–37.
- Loginova, N. V., T. V. Koval’Chuk, Y. V. Faletrov, Y. S. Halauko, N. P. Osipovich, G. I. Polozov, R. A. Zheldakova, A. T. Gres, A. S. Halauko, I. I. Azarko, V. M. Shkumatov, and O. I. Shadyro. 2011. “Redox-Active Metal(II) Complexes of Sterically Hindered Phenolic Ligands: Antibacterial Activity and Reduction of Cytochrome c. Part II. Metal(II) Complexes of o-Diphenol Derivatives of Thioglycolic Acid.” *Polyhedron* 30(15):2581–91.
- Lok, Chun Nam, Taotao Zou, Jing Jing Zhang, Iris Wing Shan Lin, and Chi Ming Che. 2014. “Controlled-Release Systems for Metal-Based Nanomedicine: Encapsulated/Self-Assembled Nanoparticles of Anticancer Gold(III)/Platinum(II) Complexes and Antimicrobial Silver Nanoparticles.” *Advanced Materials* 26(31):5550–57.
- López-Otín, Carlos, Federico Pietrocola, David Roiz-Valle, Lorenzo Galluzzi, and Guido Kroemer. 2023. “Meta-Hallmarks of Aging and Cancer.” *Cell Metabolism* 35(1):12–35.
- Lucaciu, Roxana Liana, Adriana Corina Hangan, Bogdan Sevastre, and Luminița Simona Oprean. 2022. “Metallo-Drugs in Cancer Therapy: Past, Present and Future.” *Molecules* 27(19):1–30.
- Mafukidze, Donovan M., Azole Sindelo, and T. Nyokong. 2019. “Spectroscopic Characterization and Photodynamic Antimicrobial Chemotherapy of Phthalocyanine-Silver Triangular Nanoprism Conjugates When Supported on Asymmetric Polymer Membranes.” *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy* 1–40.
- Malik, Jonaid Ahmad, Sakeel Ahmed, Bisma Jan, Onur Bender, Turki Al Hagbani, Aali Alqarni, and Sirajudheen Anwar. 2022. “Drugs Repurposed: An Advanced Step towards the Treatment of Breast Cancer and Associated Challenges.” *Biomedicine and*

Pharmacotherapy 145:112375.

- Mallandur, Bhargava K., Gururaja Rangaiah, and Nanishankar V. Harohally. 2017. "Synthesis and Antimicrobial Activity of Schiff Bases Derived from 2-Chloro Quinoline-3-Carbaldehyde and Its Derivatives Incorporating 7-Methyl-2-Propyl-3H-Benzoimidazole-5-Carboxylic Acid Hydrazide." *Synthetic Communications* 47(11):1065–70.
- Manimegalai, Sengani, Vijayarangan Devi Rajeswari, Ranganathan Parameswari, Marcello Nicoletti, Saud Alarifi, and Marimuthu Govindarajan. 2022. "Green Synthesis, Characterization and Biological Activity of Solanum Trilobatum-Mediated Silver Nanoparticles." *Saudi Journal of Biological Sciences* 29(4):2131–37.
- Manvatkar, V. D., R. Y. Patle, P. H. Meshram, and R. S. Dongre. 2023. "Azomethine-Functionalized Organic–Inorganic Framework: An Overview." *Chemical Papers* 77(10):5641–62.
- Marella, Akranth, Om Prakash Tanwar, Rikta Saha, Mohammad Rahmat Ali, Sandeep Srivastava, Mymoona Akhter, and Mohammad Shaquiquzzaman. 2013. "Quinoline : A Versatile Heterocyclic." *Saudi Pharmaceutical Journal* 21(1):1–12.
- Maret, Wolfgang. 2016. "The Biological Inorganic Chemistry of Zinc Ions *."
- Marinova, Petja Emilova and Kristina Dimova Tamahkyarova. 2024. "Synthesis and Biological Activities of Some Metal Complexes of 2-Thiouracil and Its Derivatives: A Review." *Compounds* 4(1):186–213.
- Al Mashud, Md Abdullah, Md Moinuzzaman, Md Shamim Hossain, Sabbir Ahmed, Galib Ahsan, Abu Reza, Robayet Bin Anwar Ratul, Md Helal Uddin, Md Abdul Momin, and Mohammad Abu Hena Mostofa Jamal. 2022. "Green Synthesis of Silver Nanoparticles Using Cinnamomum Tamala (Tejpata) Leaf and Their Potential Application to Control Multidrug Resistant Pseudomonas Aeruginosa Isolated from Hospital Drainage Water." *Heliyon* 8(7):e09920.
- Masoud, M. S., A. A. Soayed, S. A. Almesmari, and R. M. I. Elsamra. 2021. "New Mixed-Ligand Complexes of Cytosine and Its Silver Nanoparticles: Spectral, Analytical, Theoretical and Biological Activity Studies." *Journal of Inorganic and Organometallic Polymers and Materials* 31(7):2842–58.
- Mathuber, Marlene, Hemma Schue, Orsolya Do, Claudia Karnthaler, E. A. Enyedy, Petra He, Bernhard K. Keppler, and Christian R. Kowol. 2020. "Improving the Stability of EGFR

- Inhibitor Cobalt(III) Prodrugs.” *Inorganic Chemistry* 59:17794–810.
- Matlou, Gauta Gold and Tebello Nyokong. 2020a. “Photophysico-Chemical Properties and Photoinactivation of Staphylococcus Aureus Using Zinc Phthalocyanines Linked Silver Nanoparticles Conjugates.” *Dyes and Pigments* 176:1–11.
- Matlou, Gauta Gold and Tebello Nyokong. 2020b. “Photophysico-Chemical Properties and Photoinactivation of Staphylococcus Aureus Using Zinc Phthalocyanines Linked Silver Nanoparticles Conjugates.” *Dyes and Pigments* 108237.
- Mehata, Mohan Singh. 2021. “Green Route Synthesis of Silver Nanoparticles Using Plants/Ginger Extracts with Enhanced Surface Plasmon Resonance and Degradation of Textile Dye.” *Materials Science and Engineering: B* 273(January):115418.
- Melkamu, Walelign Wubet and Legesse Terefe Bitew. 2021. “Green Synthesis of Silver Nanoparticles Using Hagenia Abyssinica (Bruce) J.F. Gmel Plant Leaf Extract and Their Antibacterial and Anti-Oxidant Activities.” *Heliyon* 7(11):e08459.
- Meng, Xuan-Yu, Hong-Xing Zhang, Mihaly Mezei, and Meng Cui. 2011. “Molecular Docking: A Powerful Approach for Structure-Based Drug Discovery. Current Computer-Aided Drug Design.” *Current Computer Aided Drug Design* 7(2):146–57.
- Meva, François Eya’ane, Marcelle Loretta Segnou, Cecile Okalla Ebongue, Agnes Antoinette Ntomba, Philippe Belle Ebanda Kedi, Vandi Deli, Marie Annie Etoh, and Emmanuel Mpondo Mpondo. 2016. “Spectroscopic Synthetic Optimizations Monitoring of Silver Nanoparticles Formation from Megaphrynium Macrostachyum Leaf Extract.” *Revista Brasileira de Farmacognosia* 26(5):640–46.
- Mishra, Monali P., Sibanarayan Rath, Shasank S. Swain, Goutam Ghosh, Debajyoti Das, and Rabindra N. Padhy. 2017. “In Vitro Antibacterial Activity of Crude Extracts of 9 Selected Medicinal Plants against UTI Causing MDR Bacteria.” *Journal of King Saud University - Science* 29(1):84–95.
- Miu, Bogdan Andrei and Anca Dinischiotu. 2022. “New Green Approaches in Nanoparticles Synthesis: An Overview.” *Molecules* 27(19).
- Mohammad, Somaia, Sahar Ahmed, Abdel Badawi, and D. El-Desouki. 2015. “Activated Carbon Derived from Egyptian Banana Peels for Removal of Cadmium from Water.” *Journal of Applied Life Sciences International* 3(2):77–88.
- Molyneux, Philip. 2004. “The Use of the Stable Free Radical Diphenylpicryl- Hydrazyl (

- DPPH) for Estimating Antioxidant Activity.” *Journal Science and Technology* 26(211–219).
- Mondal, Suresh K., Sourav Chakraborty, Sounik Manna, and Santi M. Mandal. 2024. “Pharmaceutics Antimicrobial Nanoparticles : Current Landscape and Future Challenges.” *RSC Pharmaceutics* 1:388–402.
- Morales-avila, Enrique, Guillermina Ferro-flores, Blanca E. Ocampo-garcía, Gustavo López-téllez, Johnny López-ortega, Diana G. Rogel-ayala, and Diego Sánchez-padilla. 2017. “Antibacterial Efficacy of Gold and Silver Nanoparticles Functionalized with the Ubiquicidin (29 – 41) Antimicrobial Peptide.” *Journal of Nanomaterials* 2017:1–10.
- Moses, Colette, Benjamin Garcia-Bloj, Alan R. Harvey, and Pilar Blancafort. 2018. “Hallmarks of Cancer: The CRISPR Generation.” *European Journal of Cancer* 93:10–18.
- Mostafa, Mai A., Mostafa M. Ismail, Jehan M. Morsy, and Hany M. Hassanin. 2023. “Activities of Chitosan Schiff Bases Bearing Quinolinone or Pyranoquinolinone and Their Silver Nanoparticles Derivatives.” *Polymer Bulletin* 80(4):4035–59.
- Mourdikoudis, Stefanos and Roger M. Pallares. 2018. “Characterization Techniques for Nanoparticles : Comparison and Complementarity upon Studying.” *Nanoscale* 10:12871–934.
- Murray, Christopher JL, Kevin Shunji Ikuta, Fablina Sharara, Lucien Swetschinski, Gisela Robles Aguilar,. 2022. “Global Burden of Bacterial Antimicrobial Resistance in 2019: A Systematic Analysis.” *The Lancet* 399(10325):629–655.
- Muteeb, Ghazala, Tabish Rehman, Moayad Shahwan, and Mohammad Aatif. 2023. “Origin of Antibiotics and Antibiotic Resistance , and Their Impacts on Drug Development : A Narrative Review.” *Pharmaceutics Review* 16(1515):1–54.
- Naganagowda, Gadada, Reinout Meijboom, and Amorn Petsom. 2014. “Synthesis and Antimicrobial Activity of New Schiff Base Compounds Containing 2-Hydroxy-4-Pentadecylbenzaldehyde Moiety.” *Advanced in Chemistry* 2014:1–10.
- Ndagi, Umar, Ndumiso Mhlongo, and Mahmoud E. Soliman. 2017. “Metal Complexes in Cancer Therapy – An Update from Drug Design Perspective.” *Drug Design, Development and Therapy* 11:599–616.
- Nikoorazm, Mohsen, Parisa Moradi, Nourolah Noori, and Gouhar Azadi. 2021. “L-Arginine Complex of Copper on Modified Core–Shell Magnetic Nanoparticles as Reusable and

- Organic–Inorganic Hybrid Nanocatalyst for the Chemoselective Oxidation of Organosulfur Compounds.” *Journal of the Iranian Chemical Society* 18(2):467–78.
- Nurul, Ardiyah, Fitri Marzaman, Tri Puspita Roska, Sartini Sartini, Rifka Nurul Utami, Sulistiawati Sulistiawati, Cindy Kristina Enggi, Marianti A. Manggau, Latifah Rahman, Venkatram Prasad Shastri, and Andi Dian Permana. 2023. “Recent Advances in Pharmaceutical Approaches of Antimicrobial Agents for Selective Delivery in Various.”
- Nwobodo, David Chinemerem, Malachy Chigozie Ugwu, Clement Oliseloke Anie, Mushtak T. S. Al-Ouqaili, Joseph Chinedu Ikem, Uchenna Victor Chigozie, and Morteza Saki. 2022. “Antibiotic Resistance: The Challenges and Some Emerging Strategies for Tackling a Global Menace.” *Journal of Clinical Laboratory Analysis* 36(9):1–10.
- Odularu, Ayodele Temidayo. 2023. “Ease to Challenges in Achieving Successful Synthesized Schiff Base, Chirality, and Application as Antibacterial Agent.” *BioMed Research International* 2023(3).
- Oliver, Ana López-serrano, Marie-noële Croteau, Tasha L. Stoiber, Mila Tejamaya, Isabella Römer, Jamie R. Lead, and Samuel N. Luoma. 2014. “Does Water Chemistry Affect the Dietary Uptake and Toxicity of Silver Nanoparticles by the Freshwater Snail *Lymnaea stagnalis* ?” *Environmental Pollution* 189:87–91.
- Oves, Mohammad, Mohd Ahmar Rauf, Mohammad Aslam, Huda A. Qari, Hana Sonbol, Irfan Ahmad, Gaffar Sarwar Zaman, and Mohd Saeed. 2022. “Green Synthesis of Silver Nanoparticles by *Conocarpus lancifolius* Plant Extract and Their Antimicrobial and Anticancer Activities.” *Saudi Journal of Biological Sciences* 29(1):460–71.
- Pal, Indrani, Varsha P. Brahmkhatri, Swapna Bera, Dipita Bhattacharyya, Yasrib Quirishi, Anirban Bhunia, and Hanudatta S. Atreya. 2016. “Enhanced Stability and Activity of an Antimicrobial Peptide in Conjugation with Silver Nanoparticle.” *Journal of Colloid and Interface Science* 483:385–93.
- Pandit, Chetan, Arpita Roy, Suresh Ghotekar, Ameer Khusro, Mohammad Nazmul Islam, Talha Bin Emran, Siok Ee Lam, Mayeen Uddin Khandaker, and David Andrew Bradley. 2022. “Biological Agents for Synthesis of Nanoparticles and Their Applications.” *Journal of King Saud University - Science* 34(3):101869.
- Paramasivam, Gokul, Vishnu Vardhan Palem, Thanigaivel Sundaram, Vickram Sundaram, Somasundaram Chandra Kishore, and Stefano Bellucci. 2021. “Nanomaterials: Synthesis

- and Applications in Theranostics.” *Nanomaterials* 11(12).
- Patil, Siddappa A, Vishal Kandathil, Anjali Sobha, Sasidhar B. Somappa, Max R. Feldman, Alejandro Bugarin, and Shivaputra A Patil. 2022. “Comprehensive Review on Medicinal Applications of Coumarin-Derived Imine–Metal Complexes.” *Molecules*.
- Patra, Jayanta Kumar, Gitishree Das, Leonardo Fernandes Fraceto, Estefania Vangelie Ramos Campos, Maria Del Pilar Rodriguez-Torres, Laura Susana Acosta-Torres, Luis Armando Diaz-Torres, Renato Grillo, Mallappa Kumara Swamy, Shivesh Sharma, Solomon Habtemariam, and Han Seung Shin. 2018. “Nano Based Drug Delivery Systems: Recent Developments and Future Prospects 10 Technology 1007 Nanotechnology 03 Chemical Sciences 0306 Physical Chemistry (Incl. Structural) 03 Chemical Sciences 0303 Macromolecular and Materials Chemistry 11 Medical and He.” *Journal of Nanobiotechnology* 16(71):1–33.
- Pellei, Maura, Fabio Del, Marina Porchia, and Carlo Santini. 2021. “Zinc Coordination Complexes as Anticancer Agents.” *Coordination Chemistry Reviews* 445:214088.
- Pinheiro, Adriana Castro, Ianka Jacondino Nunes, Wesley Vieira Ferreira, Paula Pellenz Tomasini, Cristiano Trindade, Carolina Cristóvão Martins, Ethel Antunes Wilhelm, Robson da Silva Oliboni, Paulo Augusto Netz, Rafael Stieler, Osvaldo de Lazaro Casagrande, and Jenifer Saffi. 2023. “Antioxidant and Anticancer Potential of the New Cu(II) Complexes Bearing Imine-Phenolate Ligands with Pendant Amine N-Donor Groups.” *Pharmaceutics* 15(2).
- Polani, Shlomi, Semyon Melamed, Luba Burlaka, Fernando De La Vega, and David Zitoun. 2017. “Large-Scale Synthesis of Polyhedral Ag Nanoparticles for Printed Electronics.” *RSC Advances* 7(86):54326–31.
- Prashantha, A. G. and A. S. Jagadisha. 2023. “Recent Review on Synthesis, Spectral Studies, Versatile Applications of Azo Dyes and Its Metal Complexes.” *Results in Chemistry* 5(March):100890.
- Prestinaci, Francesca, Patrizio Pezzotti, and Annalisa Pantosti. 2015. “Antimicrobial Resistance: A Global Multifaceted Phenomenon.” *Pathogens and Global Health* 109(7):309–18.
- Pucelik, Barbara, Agata Barzowska, Adam Sułek, Mateusz Werłos, and Janusz M. Dąbrowski. 2024. “Refining Antimicrobial Photodynamic Therapy : Effect of Charge Distribution

- and Central Metal Ion in Fluorinated Porphyrins on Effective Control of Planktonic and Biofilm Bacterial Forms.” *Photochemical & Photobiological Sciences* 23(3):539–60.
- Purohit, Ayushi, Radheshyam Sharma, R. Shiv Ramakrishnan, Stuti Sharma, Ashish Kumar, Devendra Jain, Himmat S. Kushwaha, and Elina Maharjan. 2022. “Biogenic Synthesis of Silver Nanoparticles (AgNPs) Using Aqueous Leaf Extract of *Buchanania Lanzas* Spreng and Evaluation of Their Antifungal Activity against Phytopathogenic Fungi.” *Bioinorganic Chemistry and Applications* 2022.
- Quevedo, Ana C., Laura-jayne A. Ellis, Iseult Lynch, and Eugenia Valsami-jones. 2021. “Mechanisms of Silver Nanoparticle Uptake by Embryonic Zebrafish Cells.”
- Rajkumar, K. S., N. Kanipandian, and R. Thirumurugan. 2016. “Toxicity Assessment on Haematology, Biochemical and Histopathological Alterations of Silver Nanoparticles-Exposed Freshwater Fish *Labeo Rohita*.” *Applied Nanoscience* 6:19–29.
- Ram, Sushant and Vishnu Shankar Sinha. 2015. “Qualitative Phytochemical Analysis of Some Plants Use to Cure Malaria in Kolhan Region, of Jharkhand, India.” ~ 60 ~ *Journal of Medicinal Plants Studies* 3(6):60–62.
- Rani, Kanika, Nisha Devi, Prakash Banakar, Pushpa Kharb, and Prashant Kaushik. 2022. “Nematicidal Potential of Green Silver Nanoparticles Synthesized Using Aqueous Root Extract of *Glycyrrhiza Glabra*.” *Nanomaterials* 12(17).
- Rasheed, Wasia, Muhammad Raza Shah, Samina Perveen, Shakil Ahmed, and Sami Uzzaman. 2018. “Revelation of Susceptibility Differences Due to Hg(II) Accumulation in *Streptococcus Pyogenes* against CX-AgNPs and Cefixime by Atomic Force Microscopy.” *Ecotoxicology and Environmental Safety* 147(August 2017):9–16.
- RBenjamin, Sun, Rowe, 2005 Tang et al., and Rowe. 2008. “基因的改变” NIH Public Access.” *Bone* 23(1):1–7.
- Reddy, P. Rabindra and A. Mohan Reddy. 1996. “Synthesis and Characterization of Mixed Ligand Complexes of Zinc(II) with Cytidine and Amino Acids.” *Proceedings of the Indian Academy of Sciences - Chemical Sciences* 108(3):272.
- Reddy, V. Prakash. 2023. “Oxidative Stress in Health and Disease.” *Biomedicines* 11(11):2925 (1-17).
- Reygaert, Wanda C. 2018. “An Overview of the Antimicrobial Resistance Mechanisms of Bacteria.” 4(June):482–501.

- Reza, Seid, Fuat Topuz, Dagmara Bajer, Zahra Mohebi, Maryam Shafieioun, Hajar Heydari, Shruti Rawal, Anbazhagan Sathiyaseelan, Myeong-hyeon Wang, Rubiya Khursheed, M. H. Enayati, and Hadis Rostamabadi. 2023. "Biomedicine & Pharmacotherapy Metal Nanoparticles and Carbohydrate Polymers Team up to Improve Biomedical Outcomes." *Biomedicine & Pharmacotherapy* 168(August):115695.
- Riaz, Muhammad, Uzma Sharafat, Nafeesa Zahid, Muhammad Ismail, Jeongwon Park, Bashir Ahmad, Neelum Rashid, Muhammad Fahim, Muhammad Imran, and Aisha Tabassum. 2022. "Synthesis of Biogenic Silver Nanocatalyst and Their Antibacterial and Organic Pollutants Reduction Ability." *ACS Omega* 7(17):14723–34.
- Rimsza, Jessica M., Eric G. Sorte, and Todd M. Alam. 2019. "Hydration and Hydroxylation of MgO in Solution: NMR Identification of Proton-Containing Intermediate Phases." *ACS Omega* 4(1):1033–44.
- Saiyed, Tanzimjahan A., Jerry O. Adeyemi, Moganavelli Singh, Sunday N. Okafor, and Damian C. Onwudiwe. 2023. "Synthesis, Characterization, and Biological Evaluation of Some Metal Complexes Containing N and S Donor Atoms." *Results in Chemistry* 6(April):101052.
- Salam, M. A., Al-Amin, M. Y., Salam, M. T., Pawar, J. S., Akhter, N., Rabaan, A. A., & Alqumber, M. A. A. (2023). Antimicrobial Resistance: A Growing Serious Threat for Global Public Health. *Healthcare*, 11(13), 1–20
- Salleh, Atiqah, Ruth Naomi, Nike Dewi Utami, Abdul Wahab Mohammad, Ebrahim Mahmoudi, Norlaila Mustafa, and Mh Busra Fauzi. 2020. "The Potential of Silver Nanoparticles for Antiviral and Antibacterial Applications : A Mechanism of Action." *Nanomaterials Review* 10(1566):1–20.
- Sanità, Gennaro, Barbara Carrese, and Annalisa Lamberti. 2020. "Nanoparticle Surface Functionalization: How to Improve Biocompatibility and Cellular Internalization." *Frontiers in Molecular Biosciences* 7(November).
- Santos, Joana A. and Meindert H. Lamers. 2020. "Novel Antibiotics Targeting Bacterial Replicative DNA Polymerases." *Antibiotics* 9(776):1–14.
- Sayed, Hanaa M., Mahmoud A. Ramadan, Heba H. Salem, and Marwa A. A. Fayed. 2023. "Phytoconstituent Isolation and Cytotoxicity Evaluation of the Egyptian Cassia Occidentalis L. Possessing Selective Activity against Lung Carcinoma." *Journal of*

Chemistry 2023.

- Selgelid, Michael J. 2020. *Ethics and Drug Resistance : Collective Responsibility for Global Public Health*.
- Senguttuvan, Jamuna, Subramaniam Paulsamy, and Krishnamoorthy Karthika. 2014. "Phytochemical Analysis and Evaluation of Leaf and Root Parts of the Medicinal Herb, *Hypochaeris Radicata* L. for in Vitro Antioxidant Activities." *Asian Pacific Journal of Tropical Biomedicine* 4(Suppl 1):S359–67.
- Sevgi, Fatih, Ugur Bagkesici, Ahmed Nuri Kursunlu, and Ersin Guler. 2017. "AC SC." *Journal of Molecular Structure* (Iii).
- Shahzadi, Tayyaba, Shazia Iqbal, Tauheeda Riaz, and Maria Zaib. 2022. "A Comparative Study Based on Localized Surface Plasmon Resonance Optical Characteristics of Green Synthesized Nanoparticles towards Spectrophotometric Determination of Cupric Ions." *Journal of Taibah University for Science* 16(1):912–22.
- Shaikh, Wasim Akram, Sukalyan Chakraborty, Gary Owens, and Rafique Ul Islam. 2021. *A Review of the Phytochemical Mediated Synthesis of AgNP (Silver Nanoparticle): The Wonder Particle of the Past Decade*. Vol. 11. Springer International Publishing.
- Shakir, M., Summaiya Hanif, Mohd Asif Sherwani, Owais Mohammad, and Saud I. Al-Resayes. 2015. "Pharmacologically Significant Complexes of Mn(II), Co(II), Ni(II), Cu(II) and Zn(II) of Novel Schiff Base Ligand, (E)-N-(Furan-2-Yl Methylene) Quinolin-8-Amine: Synthesis, Spectral, XRD, SEM, Antimicrobial, Antioxidant and in Vitro Cytotoxic Studies." *Journal of Molecular Structure* 1092:143–59.
- Shankar, Shiv and Jong-whan Rhim. 2015. "Amino Acid Mediated Synthesis of Silver Nanoparticles and Preparation of Antimicrobial Agar/Silver Nanoparticles Composite Film." *Carbohydrate Polymers* 1–19.
- Sharma, Bharti, Sudeep Shukla, Rohit Rattan, Musarrat Fatima, Mayurika Goel, Mamta Bhat, Shruti Dutta, Rakesh Kumar Ranjan, and Mamta Sharma. 2022. "Antimicrobial Agents Based on Metal Complexes : Present Situation and Future Prospects." 2022.
- Sharma, Nitin Kumar, Jyotsna Vishwakarma, Summi Rai, Taghrid S. Alomar, Najla Almasoud, and Ajaya Bhattarai. 2022. "Green Route Synthesis and Characterization Techniques of Silver Nanoparticles and Their Biological Adeptness." *ACS Omega* 7(31):27004–20.

- Shobana, Chellappan, Basuvannan Rangasamy, Devan Hemalatha, and Mathan Ramesh. 2021. "Bioaccumulation of Silver and Its Effects on Biochemical Parameters and Histological Alterations in an Indian Major Carp Labeo Rohita." *Environmental Chemistry and Ecotoxicology* 3:51–58.
- Siddiqi, Khwaja Salahuddin, Aziz ur Rahman, Tajuddin, and Azamal Husen. 2018. "Properties of Zinc Oxide Nanoparticles and Their Activity Against Microbes." *Nanoscale Research Letters* 13(141):1–13.
- Siegel, Rebecca L., Kimberly D. Miller, Nikita Sandeep, and Wagle Mbbs. 2023. "Cancer Statistics , 2023." *CA: A Cancer Journal for Clinicians* (October 2022):17–48.
- Simunkova, Miriama, Peter Lauro, Klaudia Jomova, Martin Danko, Saleh Alwasel, Ibrahim M. Alhazza, Simona Rajcaniova, Zuzana Kozovska, Lucia Kucerova, Lubomir Svorc, and Marian Valko. 2019. "Redox-Cycling and Intercalating Properties of Novel Mixed Copper(II) Complexes with Non-Steroidal Anti-Inflammatory Drugs Tolfenamic, Mefenamic and Flufenamic Acids and Phenanthroline Functionality: Structure, SOD-Mimetic Activity, Interaction with Albumi." *Journal of Inorganic Biochemistry* 1-16
- Singh, A., Kukreti, R., Saso, L., & Kukreti, S. (2019). Oxidative stress: Role and response of short guanine tracts at genomic locations. *International Journal of Molecular Sciences*, 20(17).1-22
- Singh, Harjeet, Martin F. Desimone, Shivani Pandya, Srushti Jasani, Noble George, Mohd Adnan, Abdu Aldarhami, Abdulrahman S. Bazaid, and Suliman A. Alderhami. 2023. "Revisiting the Green Synthesis of Nanoparticles: Uncovering Influences of Plant Extracts as Reducing Agents for Enhanced Synthesis Efficiency and Its Biomedical Applications." *International Journal of Nanomedicine* 18(August):4727–50.
- Skłodowski, Karol, Sylwia Joanna Chmielewska-Deptuła, Ewelina Piktel, Przemysław Wolak, Tomasz Wollny, and Robert Bucki. 2023. "Metallic Nanosystems in the Development of Antimicrobial Strategies with High Antimicrobial Activity and High Biocompatibility." *International Journal of Molecular Sciences* 24(3).
- Smith, Matthew J., Fan Yang, Alexander Griffiths, Alexander Morrell, Sarah J. Chapple, Richard C. M. Siow, Theodora Stewart, Wolfgang Maret, and Giovanni E. Mann. 2023. "Redox Biology Redox and Metal Profiles in Human Coronary Endothelial and Smooth Muscle Cells under Hyperoxia , Physiological Normoxia and Hypoxia : Effects of NRF2

- Signaling on Intracellular Zinc.” *Redox Biology* 62(April):102712.
- Smolková, Romana, Lukáš Smolko, Morgane Poupon And, and Erika Samol’ová. 2023. “Pentacoordinate Mixed-Ligand Zn (II) Complexes With.” *Crystals* 13(Ii):1–18.
- Sodhi, Ravinderpal Kour and Satya Paul. 2019. “Metal Complexes in Medicine : An Overview and Update from Drug Design Perspective.” 14(1).
- Some, Sudip, Onur Bulut, Kinkar Biswas, Anoop Kumar, Anupam Roy, Ipsita Kumar Sen, Amitava Mandal, Octavio L. Franco, İkbāl Agah İnce, Kartik Neog, Sandip Das, Sayantan Pradhan, Subhadeep Dutta, Debjoy Bhattacharjya, Soumen Saha, Pradeep K. Das Mohapatra, Anil Bhumali, B. G. Unni, Ahmet Kati, Amit Kumar Mandal, M. Deniz Yilmaz, and Ismail Ocsoy. 2019. “Effect of Feed Supplementation with Biosynthesized Silver Nanoparticles Using Leaf Extract of *Morus Indica* L. V1 on *Bombyx Mori* L. (Lepidoptera: Bombycidae).” *Scientific Reports* 9(1):1–14.
- Somee, Leila Rezaei, Parinaz Ghadam, Ahya Abdi-Ali, Soudabeh Fallah, and Ghodratollah Panahi. 2018. “Biosynthesised AgCl NPs Using *Bacillus* Sp. 1/11 and Evaluation of Their Cytotoxic Activity and Antibacterial and Antibiofilm Effects on Multi-Drug Resistant Bacteria.” *IET Nanobiotechnology* 12(6):764–72.
- Sorrenti, Valeria, Ilaria Burò, Valeria Consoli, and Luca Vanella. 2023. “Recent Advances in Health Benefits of Bioactive Compounds from Food Wastes and By-Products: Biochemical Aspects.” *International Journal of Molecular Sciences* 24(3).
- Sovari, Sara Nasiri and Fabio Zobi. 2020. “Recent Studies on the Antimicrobial Activity of Transition Metal Complexes of Groups 6 – 12.” *MDPI Chemistry* 2:418–52.
- Stephens*, P. J. .. and F. J. Devlin. 1994. “Ab Initio Calculation of Vibrational Absorption and Circular Dichroism Spectra Using Density Functional Force Fields.” *The Journal of Physical Chemistry* 98(45):11623–27.
- Sun, Leming, Hongmei Liu, Yanqi Ye, Yang Lei, Rehmat Islam, Sumin Tan, Rongsheng Tong, Yang-Bao Miao⁴, and Lulu Ca. 2023. “Smart Nanoparticles for Cancer Therapy.” *Sun, L. (2023). Smart Nanoparticles for Cancer Therapy. April. <https://doi.org/10.1038/S41392-023-01642-X> (April):1–28.*
- Sung, Hyuna, Jacques Ferlay, Rebecca L. Siegel, Mathieu Laversanne, Isabelle Soerjomataram, Ahmedin Jemal, and Freddie Bray. 2021. “Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185

- Countries.” *CA: A Cancer Journal for Clinicians* 71(3):209–49.
- Suresh, S., S. Karthikeyan, and K. Jayamoorthy. 2016. “FTIR and Multivariate Analysis to Study the Effect of Bulk and Nano Copper Oxide on Peanut Plant Leaves.” *Journal of Science: Advanced Materials and Devices* 1(3):343–50.
- Szczyglewska, Paulina, Agnieszka Feliczak-Guzik, and Izabela Nowak. 2023. “Nanotechnology–General Aspects: A Chemical Reduction Approach to the Synthesis of Nanoparticles.” *Molecules* 28(13).
- Szunyogová, Erika, Dagmar Mudro, Katarína Györyová, and Radomíra Nemcová. 2007. “The Physicochemical and Biological Properties of Zinc (II) Complexes.” *Journal Of Thermal Analysis and Calorimetry* 88(2):355–61.
- Tang, Ka Wah Kelly, Beverley C. Millar, and John E. Moore. 2023. “Antimicrobial Resistance (AMR).” *British Journal of Biomedical Science* 80(June):1–11.
- Tapabashi, Nashwan O., Nihad I. Taha, and Marwa El-subeyhi. 2021. “Design , Microwave Assisted Synthesis of Some Schiff Bases Derivatives of Congo Red and Conventional Preparation of Their Structurally Reversed Analogous Compounds.” *International Journal of Organic Chemistry* 11:35–45.
- Tesauro, Diego. 2022. “Metal Complexes in Diagnosis and Therapy.” *Internation Journal of Molecular Science* 23(8):4377(1-4).
- Thompson, Michael W. 2022. “Regulation of Zinc - Dependent Enzymes by Metal Carrier Proteins.” *BioMetals* 35(2):187–213.
- Tim, Beata, Paulina Błaszkiwicz, and Michał Kotkowiak. 2022. “Recent Advances in Metallic Nanoparticle Assemblies for Surface-Enhanced Spectroscopy.” *International Journal of Molecular Sciences* 23(1).
- Timoshnikov, Viktor A., Olga Yu Selyutina, Nikolay E. Polyakov, Victoria Didichenko, and George J. Kontoghiorghes. 2022. “Mechanistic Insights of Chelator Complexes with Essential Transition Metals : Antioxidant / Pro-Oxidant Activity and Applications in Medicine.”
- Todorov, Lozan and Irena Kostova. 2023. “Recent Trends in the Development of Novel Metal-Based.” *Molecular Pharmaceutics* 28(1959):1–33.
- Tomić, Simonida Lj and Jovana S. Vuković. 2022. “Antimicrobial Activity of Silver, Copper, and Zinc Ions/Poly(Acrylate/Itaconic Acid) Hydrogel Matrices.” *Inorganics* 10(3):1–12.

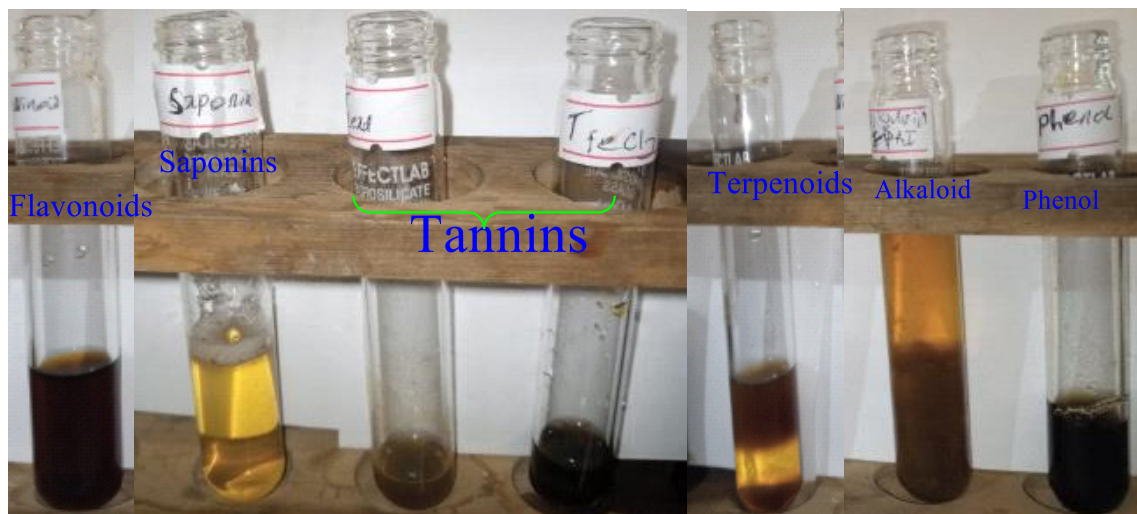
- Trang, Tran Thu, Thu Ha, Nguyen Van Dang, and Thi Nga. 2024. "Revealing the High Efficiency of Fluorescence Quenching of Rhodamine B by Triangular Silver Nanodisks Due to the Inner Filter Effect Mechanism." *RSC Advances* 14:9538–46.
- Turan, Nevin and Memet Sekerci. 2010. "Synthesis , Characterization and Thermal Behavior of Some Zn(II) Complexes with Ligands Having 1 , 3 , 4-Thiadiazole Moieties." *Heteroatom Chemistry* 21(March):13–23.
- Turan, Nevin and Memet Şekerci. 2010. "Synthesis, Characterization and Thermal Behavior of Some Zn(II) Complexes with Ligands Having 1,3,4-Thiadiazole Moieties." *Heteroatom Chemistry* 21(1):14–23.
- Turner, Raymond J. 2017. "Metal-Based Antimicrobial Strategies." 10:1062–65.
- Uddin, Tanvir Mahtab, Arka Jyoti Chakraborty, Ameer Khusro, BM Redwan Matin Zidan, Saikat Mitra, Talha Bin Emran, Kuldeep Dhama, Md Kamal Hossain Ripon, Márió Gajdács, Muhammad Umar Khayam Sahibzada, Md Jamal Hossain, and Niranjana Koirala. 2021. "Antibiotic Resistance in Microbes: History, Mechanisms, Therapeutic Strategies and Future Prospects." *Journal of Infection and Public Health* 14(12):1750–66.
- Van Der Westhuizen, D., Bezuidenhout, D. I., & Munro, O. Q. (2021). Cancer molecular biology and strategies for the design of cytotoxic gold(i) and gold(iii) complexes: A tutorial review. *Dalton Transactions*, 50(47), 17413–17437
- Venkatesh, Ganesan, Palanisamy Vennila, Savas Kaya, Samia Ben Ahmed, Paramasivam Sumathi, Vadivel Siva, Premkumar Rajendran, and Chennapan Kamal. 2024. "Synthesis and Spectroscopic Characterization of Schiff Base Metal Complexes, Biological Activity, and Molecular Docking Studies." *ACS Omega*.
- Venugopal, K., H. A. Rather, K. Rajagopal, M. P. Shanthi, K. Sheriff, M. Illiyas, R. A. Rather, E. Manikandan, S. Uvarajan, M. Bhaskar, and M. Maaza. 2017. "Synthesis of Silver Nanoparticles (Ag NPs) for Anticancer Activities (MCF 7 Breast and A549 Lung Cell Lines) of the Crude Extract of Syzygium Aromaticum." *Journal of Photochemistry and Photobiology B: Biology* 167(January):282–89.
- Waarts, Michael R., Aaron J. Stonestrom, Young C. Park, and Ross L. Levine. 2022. "Targeting Mutations in Cancer." *Journal of Clinical Investigation* 132(8):1–11.
- Wan Mat Khalir, Wan Khaima Azira, Kamyar Shameli, Seyed Davoud Jazayeri, Nor Azizi Othman, Nurfatehah Wahyuni Che Jusoh, and Norazian Mohd Hassan. 2020.

- “Biosynthesized Silver Nanoparticles by Aqueous Stem Extract of *Entada Spiralis* and Screening of Their Biomedical Activity.” *Frontiers in Chemistry* 8(August):1–15.
- Wang, Caixia, Liangxin Fan, Zhenliang Pan, Sufang Fan, Lijun Shi, Xu Li, Jinfang Zhao, Lulu Wu, Guoyu Yang, and Cuilian Xu. 2022. “Synthesis of Novel Indole Schiff Base Compounds and Their Antifungal Activities.” *Molecules* 27(20):1–15.
- Wang, Haolan, Ming Guo, Hudie Wei, and Yongheng Chen. 2023. “Targeting P53 Pathways: Mechanisms, Structures, and Advances in Therapy.” *Signal Transduction and Targeted Therapy* 8(1):1–35.
- Wang, Linlin, Chen Hu, and Longquan Shao. 2017. “The Antimicrobial Activity of Nanoparticles : Present Situation and Prospects for the Future.” *International Journal of Nanomedicine* 2017:12 12:1227–49.
- Wang, Minghua, Lei Zhu, Shuai Zhang, Yafei Lou, Shuangrun Zhao, Quan Tan, and Linghao He. 2021. “Sensors and Actuators : B . Chemical A Copper (II) Phthalocyanine-Based Metallo-Covalent Organic Framework Decorated with Silver Nanoparticle for Sensitively Detecting Nitric Oxide Released from Cancer Cells.” *Sensors and Actuators: B. Chemical* 338(March):129826.
- Wani, Waseem A., Sanjiv Prashar, Sheikh Shreaz, and Santiago Gómez-ruiz. 2016. “Nanostructured Materials Functionalized with Metal Complexes: In Search of Alternatives for Administering Anticancer Metallodrugs.” *Coordination Chemistry Reviews* 1–108.
- Werner, Haim and Derek LeRoith. 2022. “Hallmarks of Cancer: The Insulin-like Growth Factors Perspective.” *Frontiers in Oncology* 12(November):1–13.
- Weth, Freya R., Georgia B. Hoggarth, Anya F. Weth, Erin Paterson, Madeleine P. J. White, Swee T. Tan, Lifeng Peng, and Clint Gray. 2024. “Unlocking Hidden Potential: Advancements, Approaches, and Obstacles in Repurposing Drugs for Cancer Therapy.” *British Journal of Cancer* 130(5):703–15.
- WHO. 2019. *2019 Antibacterial Agents in Clinical Development: An Analysis of the Antibacterial Clinical Development Pipeline*. Geneva: World Health Organization; 2019. Licence: CC BY-NC-SA 3.0 IGO.
- Yamgar, Ramesh S., Y. Nivid, Satish Nalawade, Mustapha Mandewale, R. G. Atram, and Sudhir S. Sawant. 2014. “Novel Zinc (II) Complexes of Heterocyclic Ligands as

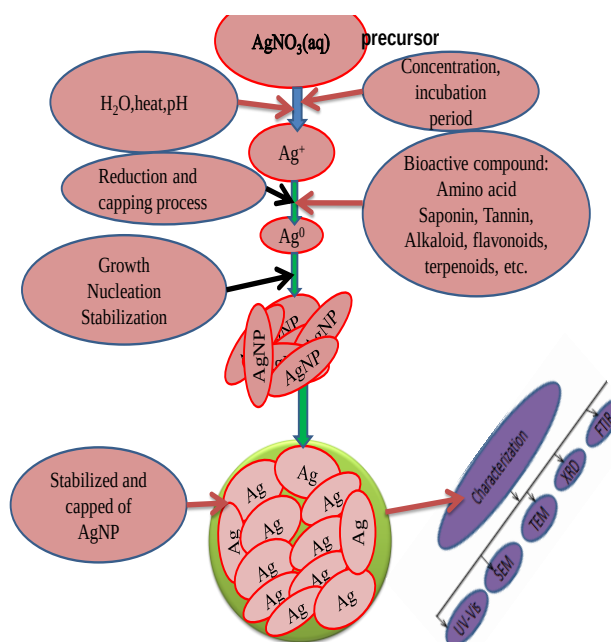
- Antimicrobial Agents: Synthesis, Characterisation, and Antimicrobial Studies.” *Bioinorganic Chemistry and Applications* 2014:1–10.
- Yang, Jingjing, Anthony Griffin, Zhe Qiang, and Jie Ren. 2022. “Organelle-Targeted Therapies: A Comprehensive Review on System Design for Enabling Precision Oncology.” *Signal Transduction and Targeted Therapy* 7(1).
- Yaqoob, Asim Ali, Hilal Ahmad, Tabassum Parveen, Akil Ahmad, Mohammad Oves, Iqbal M. I. Ismail, Huda A. Qari, Khalid Umar, and Mohamad Nasir Mohamad Ibrahim. 2020. “Recent Advances in Metal Decorated Nanomaterials and Their Various Biological Applications: A Review.” *Frontiers in Chemistry* 8(May):1–23.
- Yernale, Nagesh Gunavanthrao and Mruthyunjayaswamy Bennikallu Hire Mathada. 2020. “Preparation of Octahedral Cu(II), Co(II), Ni(II) and Zn(II) Complexes Derived from 8-Formyl-7-Hydroxy-4-Methylcoumarin: Synthesis, Characterization and Biological Study.” *Journal of Molecular Structure* 1220(Ii):128659.
- Ying, Shuaixuan, Zhenru Guan, Polycarp C. Ofoegbu, Preston Clubb, Cyren Rico, Feng He, and Jie Hong. 2022. “Green Synthesis of Nanoparticles: Current Developments and Limitations.” *Environmental Technology and Innovation* 26:102336.
- Yue, Yang, Renata Behra, Laura Sigg, Marc J. Suter, Smitha Pillai, and Kristin Schirmer. 2016. “Silver Nanoparticle–Protein Interactions in Intact Rainbow Trout Gill Cells.” *Environmental Science Nano* 3:1174–85.
- Zalevskaya, O. A. and Y. A. Gur. 2021. “Recent Studies on the Antimicrobial Activity of Copper Complexes.” 47(12):861–80.
- Zein, Randa, Wissam Sharrouf, and Kim Selting. 2020. “Physical Properties of Nanoparticles That Result in Improved Cancer Targeting.” *Journal of Oncology* 2020.
- Zewudie, Adisu Girma, Enyew Amare Zereffa, Teshome Abdo Segne, H. C. Anand. Murthy, C. R. Ravikumar, Dhanalakshmi Muniswamy, and Bayissa Bekele Binagdie. 2023. “Biosynthesis of Ag/Bentonite, ZnO/Bentonite, and Ag/ZnO/Bentonite Nanocomposites by Aqueous Leaf Extract of Hagenia Abyssinica for Antibacterial Activities.” *Reviews on Advanced Materials Science* 62(1).
- Zhang, Chunyu, Chao Xu, Xueyun Gao, and Qingqiang Yao. 2022. “Theranostics Platinum-Based Drugs for Cancer Therapy and Anti-Tumor Strategies.” *Theranostics* 12(5):2115–32.

- Zhang, Youjuan, Caiwen Zhang, and Jing Chen. 2021. "Synthesis and Crystal Structures of Two Metal-Radical Complexes." *Open Journal of Inorganic Chemistry* 11:85–94.
- Zhu, Wei, Zhanqi Wei, Chang Han, and Xisheng Weng. 2021. "Nanomaterials as Promising Theranostic Tools in Nanomedicine and Their Applications in Clinical Disease Diagnosis and Treatment." *Nanomaterials* 11(12).

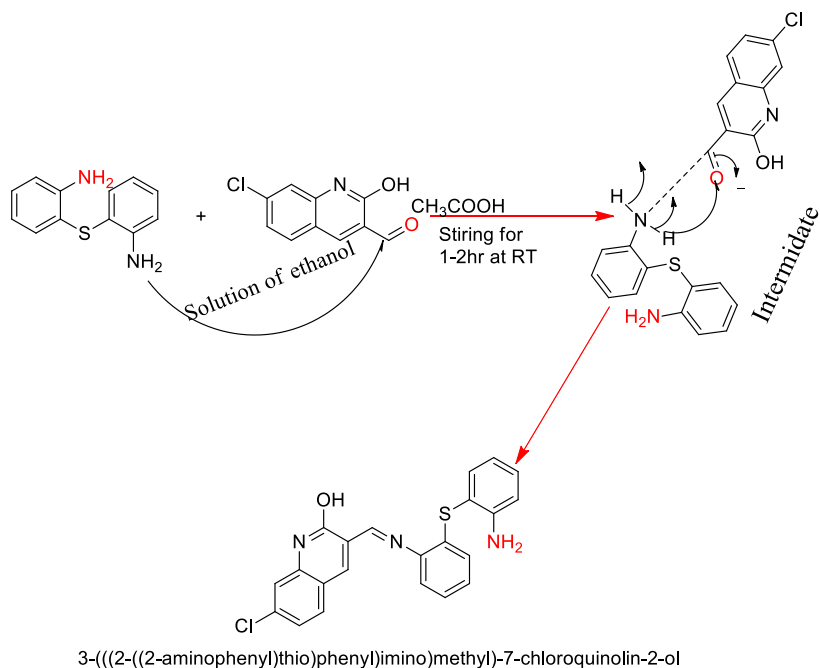
APPENDICES



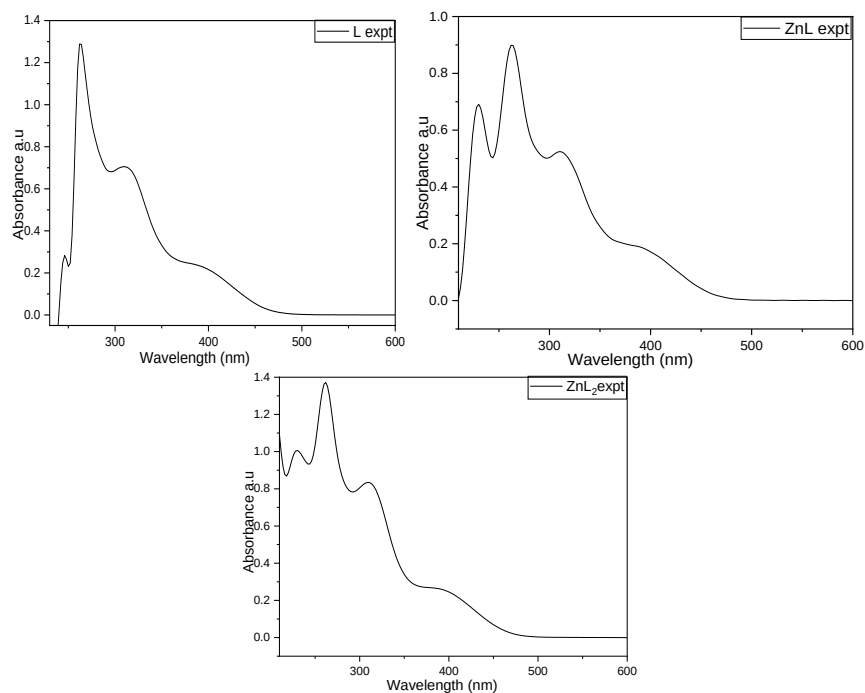
Appendix 1. Qualitative phytochemical analysis of *lippia abyssinica* leaf extract



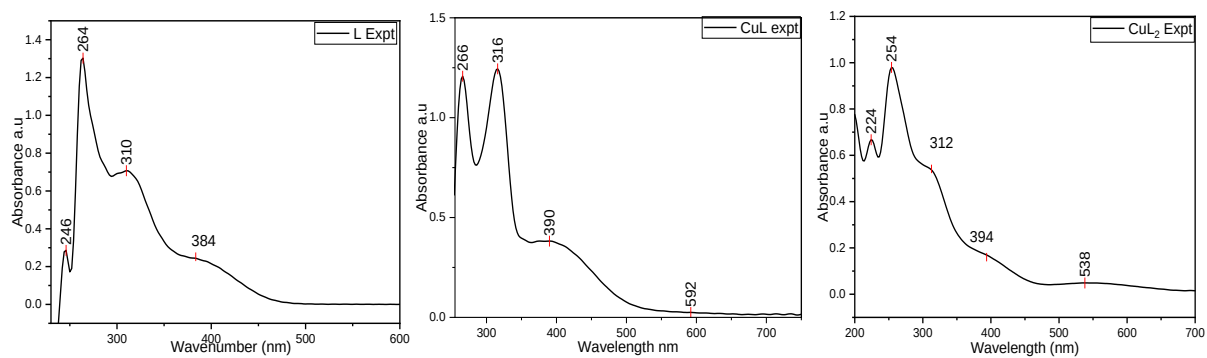
Appendix 2. Mechanism involved in LALE based synthesis and characterization of AgNPs.



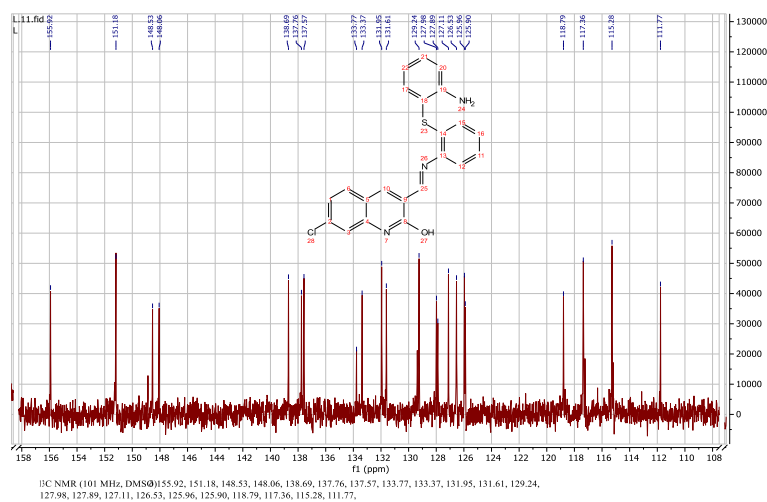
Appendix 3. The Mechanism of synthesis of imine quinoline ligand



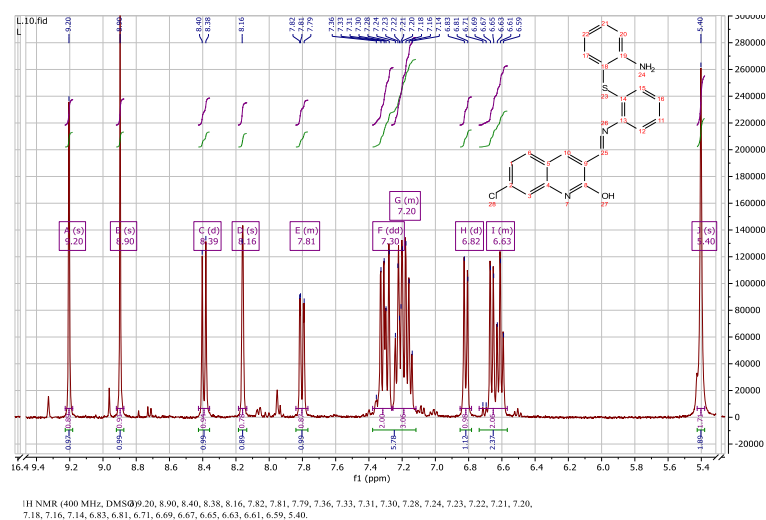
Appendix 4. Experimental UV-Vis spectra of the ligand (L expt) and its **ZnL**(expt), and **ZnL₂**(expt) complexes



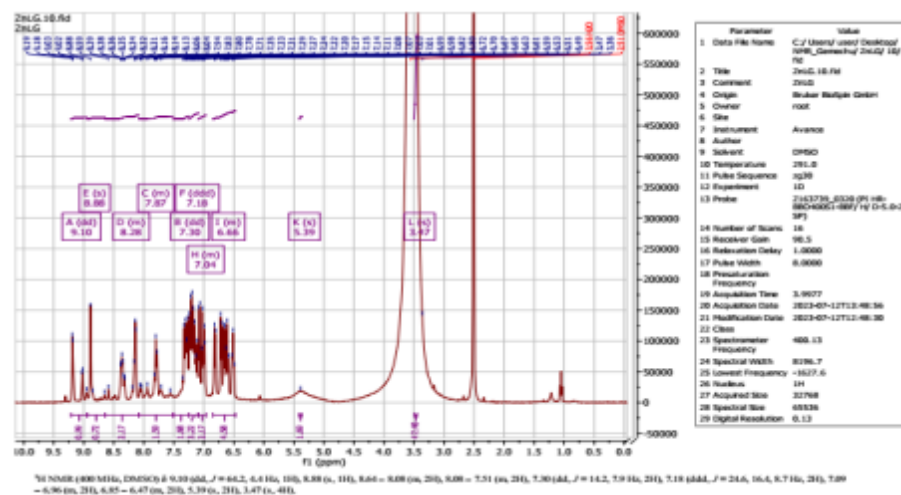
Appendix 5. UV-Vis spectra of the ligand (L), CuL, and CuL₂ complexes.



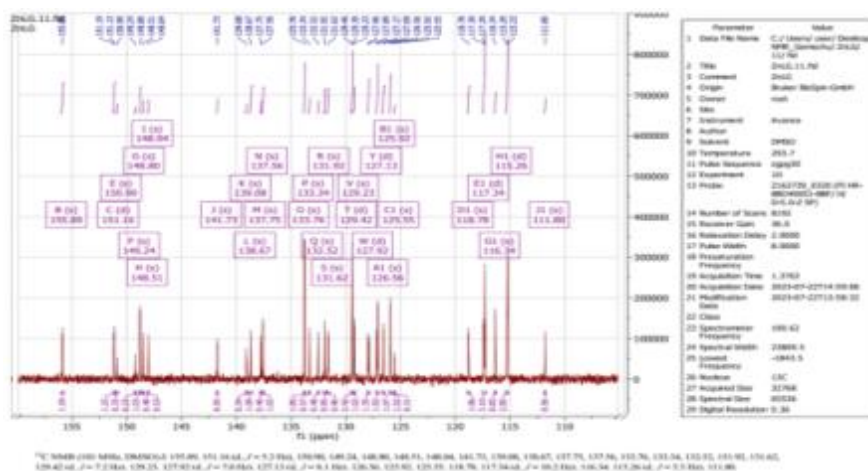
Appendix 6. The spectrum of chemical shift of ¹³C NMR of the IQL (L)



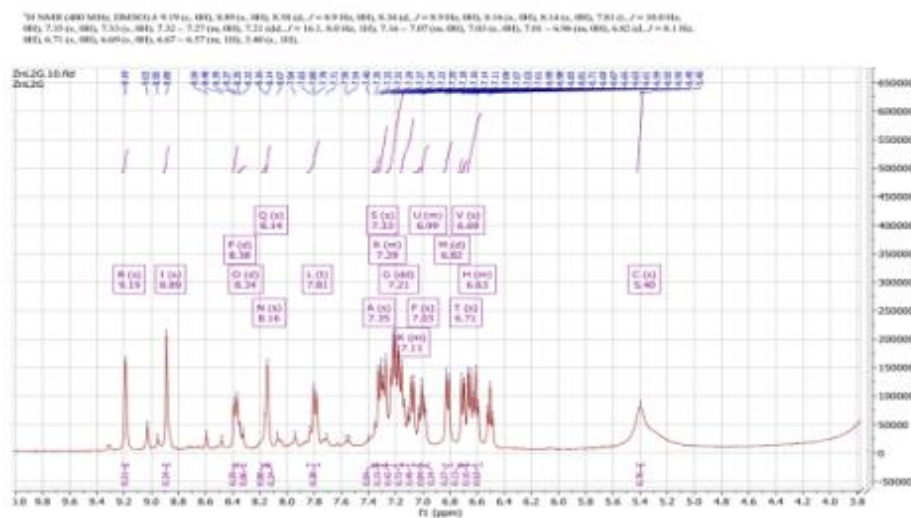
Appendix 7. The spectrum of chemical shift of ¹H NMR of the imine quinoline ligand (L)

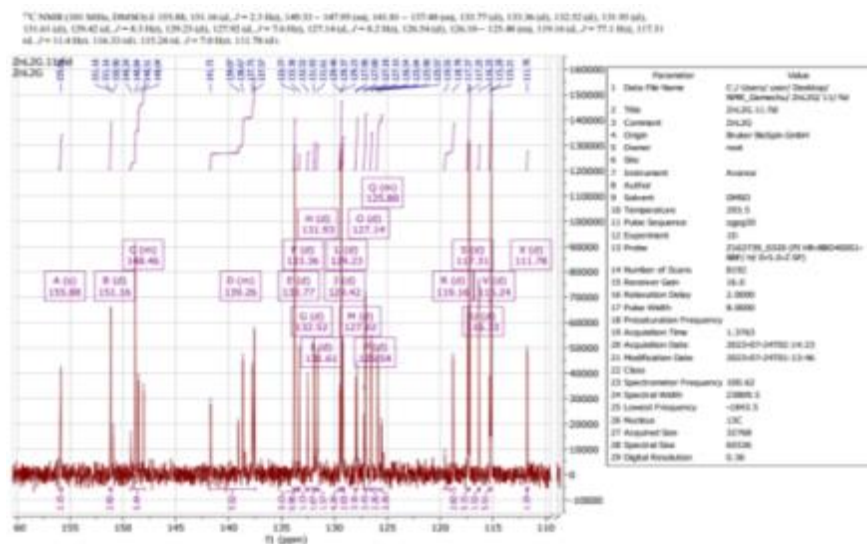


Appendix 8. The spectrum of chemical shift of ^1H NMR of the **ZnL**



Appendix 9 The spectrum of chemical shift of ^{13}C NMR of the **ZnL**





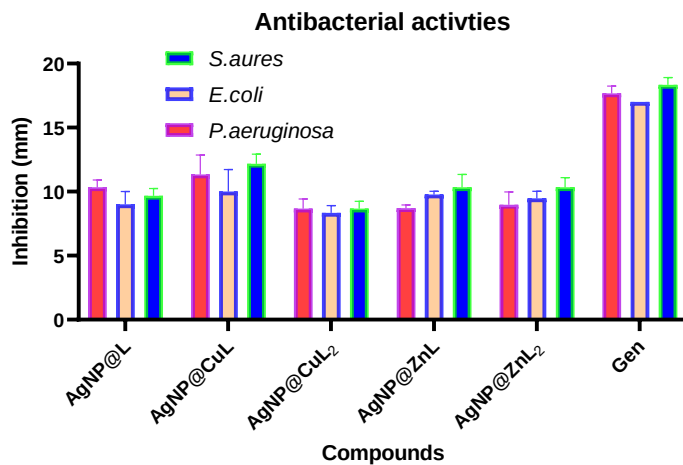
Appendix 11. The spectrum of chemical shift of ¹³C NMR of the ZnL₂



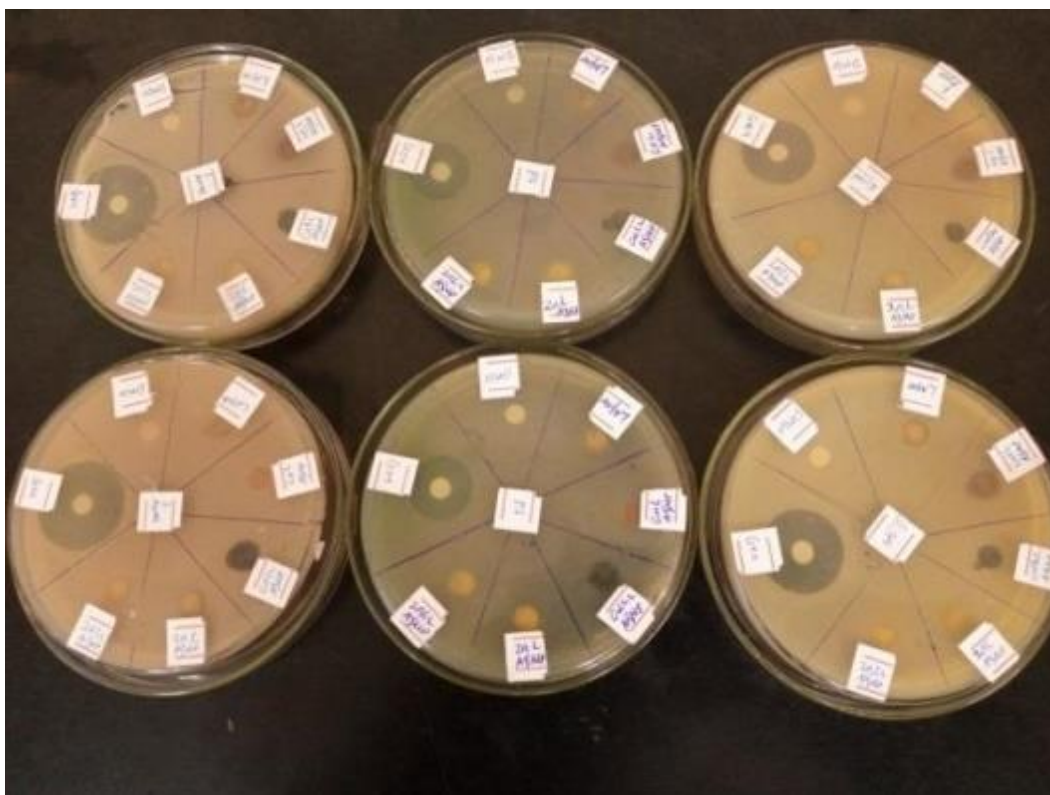
Appendix 12. Antibacterial activities of AgNPs with 62.5µg/ml of 2mM (AgNO₃) using Disc diffusion method



Appendix 13. Disc diffusion the antibacterial activities of the ligand and its Cu(II) and Zn(II) metal complex



Appendix 14. The antibacterial activities of decorated ligand, and its Cu(II) and Zn(II) with AgNPs



Appendix 15. Disc diffusion the antibacterial activities of the ligand and its Cu(II) and Zn(II) metal complex decorated AgNPs

Appendix 16. The antioxidant activities the biosynthesis of AgNPs

μg/mL	AgNP.Scv		AgNP-H.Scv		AgNP-Ph.Scv		AA.Scv	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
50	22.72	0.16	25.08	0.33	23.95	0.13	94.65	0.03
100	28.69	0.10	35.67	0.07	32.00	0.08	95.80	0.06
150	37.64	0.10	48.40	0.13	42.80	0.09	96.41	0.10
200	45.69	0.10	54.63	0.13	48.84	0.06	98.24	0.07
250	51.28	0.16	63.09	0.10	54.98	0.07	99.16	0.10

Appendix 17. The antioxidant activities the ligand, and its Cu(II) and Zn(II)

μg/mL	Ligand (L)		CuL.Scv		CuL ₂ .Scv		ZnL.Scv		ZnL ₂ .Scv		AA.Scv	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
50	23.92	0.16	26.18	0.33	25.75	0.13	24.08	0.33	25.12	0.23	94.65	0.03
100	29.89	0.10	36.77	0.07	33.80	0.08	34.67	0.07	33.16	0.16	95.80	0.06
150	38.84	0.10	49.50	0.13	44.60	0.09	47.40	0.13	43.97	0.19	96.41	0.10
200	46.89	0.10	55.73	0.13	50.64	0.06	53.63	0.13	50.01	0.09	98.24	0.07
250	52.48	0.16	64.19	0.10	56.78	0.07	62.09	0.10	56.15	0.15	99.16	0.10

Appendix 18 .The antioxidant activities of decorated ligand, and its Cu(II) and Zn(II) with AgNPs

μg/ m L	AgNP@L. Scv		AgNP@Cu L.Scv		AgNP@Cu L ₂ .Scv		AgNP@Zn L.Scv		AgNP@Zn L ₂ .Scv		AA.Scv	
	Mea n	SD	Mean	SD	Mea n	SD	Mean	SD	Mean	SD	Mean	SD
	26.1	0.2		0.7	26.7			0.9		0.9		
50	2	6	34.86	3	0	0.18	28.55	5	27.04	9	94.65	0.04
10	32.0	0.1		0.7	34.7			0.7		0.7		
0	9	9	45.26	3	5	0.12	39.14	7	35.08	3	95.80	0.06
15	41.0	0.0		0.7	45.5			1.2		1.9		
0	4	6	58.01	0	5	0.15	51.85	8	41.05	5	96.41	0.10
20	49.0	0.1		0.7	51.5			0.8		2.0		
0	9	9	64.26	1	9	0.05	58.08	0	51.92	0	98.24	0.06
25	54.6	0.2		0.7	57.7			1.6		0.9		
0	9	7	72.62	2	3	0.01	67.58	5	58.05	0	99.16	0.10

Appendix 19 B3LYP-GD3/6-31++G(d,p)/LanL2DZ/PCM/DMSO optimized geometries

<u>Ligand (L)</u>			
C	-2.181191000	0.367735000	3.130697000
C	-1.052184000	1.167641000	2.954090000
C	-0.817328000	1.806416000	1.729076000
C	-1.745709000	1.661607000	0.676349000
C	-2.870179000	0.850246000	0.861524000
C	-3.087938000	0.204258000	2.080708000
C	-2.054685000	0.434921000	-2.593914000
C	-2.451367000	1.670215000	-2.024044000
N	3.915194000	-0.642149000	1.226418000
C	3.355778000	0.510325000	1.487891000
C	-3.667297000	2.256901000	-2.400054000
C	1.979433000	0.836603000	1.194723000
C	1.209784000	-0.174439000	0.644128000
C	-4.510377000	1.639698000	-3.323501000
C	-4.126458000	0.413221000	-3.877120000
C	-2.918963000	-0.179983000	-3.519628000
C	1.484118000	2.216736000	1.383831000
N	0.297542000	2.658455000	1.569753000
S	-1.384902000	2.522300000	-0.864860000
O	4.184414000	1.410668000	2.081845000
Cl	3.761466000	-5.410304000	-0.586312000
C	3.160257000	-1.620472000	0.654222000
C	3.770858000	-2.864902000	0.354897000
N	-0.871801000	-0.193560000	-2.210862000
C	3.013067000	-3.866152000	-0.212969000
C	1.636860000	-3.699165000	-0.512407000
C	1.031231000	-2.495335000	-0.230081000
C	1.773885000	-1.435116000	0.355422000
H	1.082554000	-4.518029000	-0.956635000

H	4.821177000	-3.003056000	0.582100000
H	-0.019367000	-2.335567000	-0.456174000
H	0.167768000	-0.004719000	0.406598000
H	-2.347224000	-0.126549000	4.083017000
H	-0.333690000	1.302461000	3.756774000
H	-3.573670000	0.719481000	0.046579000
H	-3.967884000	-0.419160000	2.205995000
H	-3.944810000	3.205766000	-1.951327000
H	-5.449354000	2.105286000	-3.604309000
H	-4.769964000	-0.084643000	-4.596828000
H	-2.630365000	-1.131843000	-3.958934000
H	2.248006000	2.999207000	1.311795000
H	3.702528000	2.192446000	2.380260000
H	-0.502381000	-0.860066000	-2.875943000
H	-0.159834000	0.433615000	-1.853791000

CuL

C	-3.841722000	0.156817000	-2.087173000
C	-3.914055000	0.352002000	-0.671626000
C	-5.966047000	1.625528000	-0.944711000
O	3.509008000	-1.157711000	-1.105894000
N	2.375232000	1.858830000	4.635887000
O	1.449945000	-3.474987000	-2.046992000
Cl	0.714540000	1.005311000	-1.805730000
Cl	2.080317000	-1.315783000	-4.012390000
Cu	1.332258000	-1.191259000	-1.818854000
Cl	-7.121419000	2.112850000	-3.365755000
O	-0.937940000	-1.808931000	-2.496723000
S	0.922047000	0.668078000	2.186166000
N	0.479774000	-1.586529000	0.050106000

C	-0.728930000	-1.300643000	0.392862000
C	-1.896048000	-1.051912000	-1.901892000
N	-2.837974000	-0.569051000	-2.661103000
C	3.943883000	3.176586000	3.328059000
C	4.486789000	3.550732000	2.101192000
C	1.433438000	-1.861861000	1.073861000
C	2.133884000	-3.078617000	1.000589000
C	-4.844223000	0.709189000	-2.918764000
C	3.964890000	3.045823000	0.904580000
C	2.886444000	2.163443000	0.950204000
C	2.335497000	1.780415000	2.180553000
C	2.857746000	2.282915000	3.395164000
C	3.457089000	-2.451151000	2.922083000
C	2.768718000	-1.244034000	3.000578000
C	1.751083000	-0.926054000	2.083151000
C	-5.875072000	1.424487000	-2.344695000
C	-2.874931000	-0.198008000	0.110768000
C	-1.821327000	-0.879887000	-0.475775000
C	3.133014000	-3.377327000	1.922365000
C	-4.994480000	1.093682000	-0.125993000
H	-6.793109000	2.196294000	-0.538210000
H	-4.781398000	0.560760000	-3.990219000
H	-5.045490000	1.243848000	0.948917000
H	-2.880392000	-0.041931000	1.186977000
H	3.656739000	-4.326745000	1.859993000
H	1.891927000	-3.764072000	0.197145000
H	3.021402000	-0.524743000	3.772836000
H	4.241030000	-2.670568000	3.640785000
H	2.453867000	1.768610000	0.036163000
H	4.384986000	3.338065000	-0.052388000
H	5.323630000	4.243679000	2.081304000

H	4.363323000	3.568025000	4.252015000
H	-0.991701000	-1.345862000	1.454436000
H	1.808483000	-3.362614000	-2.950073000
H	2.536590000	2.494594000	5.405010000
H	1.436908000	1.477967000	4.624525000
H	3.953240000	-1.094649000	-1.965546000
H	0.508920000	-3.672148000	-2.165917000
H	3.832326000	-1.959459000	-0.673230000
H	-0.962110000	-1.629434000	-3.453847000

CuL₂

C	3.314169000	2.305627000	-0.460843000
C	3.490116000	3.074896000	-1.614864000
C	2.425308000	-3.322637000	0.946921000
C	2.664988000	-2.051465000	1.465405000
C	2.586201000	-1.907862000	2.899617000
N	2.084958000	-2.823241000	3.715037000
C	1.645327000	-5.667905000	1.313601000
C	1.976241000	-4.368273000	1.792485000
C	1.747598000	-4.045070000	3.179694000
C	1.162815000	-5.018161000	4.032326000
C	0.838655000	-6.267890000	3.515496000
C	1.080155000	-6.614130000	2.156939000
Cu	1.902144000	1.282870000	2.564152000
Cl	-1.804509000	-6.091308000	-1.800100000
O	-0.028671000	-0.696985000	2.972001000
S	0.159749000	3.511686000	-0.836855000
N	0.028019000	1.977552000	1.887978000
C	-0.976467000	1.423698000	1.266627000
C	-3.502629000	4.667819000	-2.338651000
C	-3.160541000	4.938710000	-3.671487000

C	-1.818176000	4.872483000	-4.083264000
C	-0.832748000	4.524581000	-3.147722000
C	-1.167054000	4.216177000	-1.811953000
C	0.016520000	4.239856000	0.796211000
C	0.033763000	3.410517000	1.955185000
C	-2.525952000	4.295217000	-1.386181000
C	-1.966877000	-0.379384000	-0.055328000
C	0.049285000	5.398634000	3.368781000
C	-2.685838000	-2.164686000	-1.644581000
N	-0.655627000	-2.346676000	1.497026000
C	-0.006757000	5.642986000	0.950951000
Cl	1.413931000	1.222649000	4.871216000
Cl	3.837213000	2.450699000	2.887018000
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C	-1.310433000	-2.699663000	0.343066000
C	-2.034643000	-1.739137000	-0.451859000
C	-0.014129000	6.220882000	2.228431000
C	-1.169502000	0.003126000	1.021777000
C	-2.614892000	-3.487739000	-2.055546000
C	-0.600980000	-1.066380000	1.810142000
C	0.086783000	4.005636000	3.232805000
C	2.593579000	1.092388000	-0.507770000
C	1.992324000	0.677736000	-1.735137000
C	2.174802000	1.473248000	-2.886865000
C	2.923263000	2.655729000	-2.832143000
C	-1.012961000	0.804220000	-2.861195000
C	-0.283204000	-0.391797000	-2.995636000
N	-2.903305000	3.942803000	-0.077847000
C	-0.613091000	-1.322649000	-4.021071000
N	0.044119000	-2.546039000	-4.134012000

C	-1.694549000	-1.002471000	-4.882008000
C	-2.408845000	0.193218000	-4.738371000
C	-2.068431000	1.114031000	-3.726880000
C	2.837610000	-0.950796000	0.528817000
N	2.512638000	0.306460000	0.685899000
S	0.997471000	-0.827757000	-1.814189000
O	3.054236000	-0.747463000	3.382050000
Cl	0.110857000	-7.472965000	4.565966000
H	-3.074757000	-3.814043000	-2.991129000
H	-0.726743000	-4.783555000	0.539237000
H	-3.193823000	-1.421086000	-2.268406000
H	-2.476081000	0.383132000	-0.654395000
H	0.082949000	5.841165000	4.368970000
H	0.164681000	3.344927000	4.101835000
H	-0.012277000	6.270637000	0.053682000
H	-0.035914000	7.310615000	2.330470000
H	0.218336000	4.459994000	-3.448698000
H	-1.538597000	5.091197000	-5.118015000
H	-3.945474000	5.214669000	-4.383043000
H	-4.550060000	4.734208000	-2.019754000
H	-1.756512000	2.070350000	0.827762000
H	0.811524000	-7.609428000	1.794406000
H	0.983002000	-4.767304000	5.080436000
H	1.818176000	-5.908273000	0.258709000
H	2.491876000	-3.491883000	-0.134787000
H	4.063519000	4.005130000	-1.559147000
H	3.737207000	2.600853000	0.506264000
H	1.705750000	1.163575000	-3.825639000
H	3.054845000	3.251482000	-3.741498000
H	-0.734215000	1.496445000	-2.062288000
H	-1.967779000	-1.713307000	-5.670786000

H	-3.234517000	0.409866000	-5.424445000
H	-2.605346000	2.060883000	-3.612752000
H	3.229832000	-1.264584000	-0.456147000
H	-3.870164000	4.184218000	0.146125000
H	-2.268409000	4.283396000	0.650882000
H	-0.002825000	-2.998922000	-5.045215000
H	0.969148000	-2.588209000	-3.701193000
H	2.718456000	-0.544051000	4.300748000
H	0.276421000	-1.502517000	3.453336000

Appendix 20. The summary of the Average of crystal grain size of synthesized compounds

Compounds	Average grain size (nm)
AgNP	26.64
L	13.28
CuL	31.47
CuL₂	11.54
ZnL	21.19
ZnL₂	16.26
L@AgNP	20.67
CuL@AgNP	20.83
CuL₂@AgNP	19.74
ZnL@AgNP	22.00
ZnL₂@AgNP	19.25

Appendix 21. The in silico analysis of ligand, CuL and ZnL using swissADME

Parameter	Ligand	ZnL	CuL
Formula	C22H16ClN3OS	C22H20Cl3N3O3SZn	C22H20Cl3CuN3O3S
MW	405.9	578.22	576.38
#Heavy atoms	28	33	33
#Aromatic heavy atoms	22	22	22
Fraction Csp3	0	0	0
#Rotatable bonds	4	3	3
#H-bond acceptors	3	4	4
#H-bond donors	2	4	4

MR	117.44	138.57	138.57
TPSA	96.8	130.26	115.26
iLOGP	3.35	0	0
XLOGP3	5.48	6.24	6.24
WLOGP	6.09	6.43	6.95
MLOGP	4.19	2.97	2.97
Silicos-IT Log P	5.42	0.19	0.17
Consensus Log P	4.9	3.16	3.27
ESOL Log S	-6.13	-7.65	-7.64
ESOL Solubility (mg/ml)	3.03E-04	1.29E-05	1.32E-05
ESOL Solubility (mol/l)	7.47E-07	2.23E-08	2.29E-08
ESOL Class	Poorly soluble	Poorly soluble	Poorly soluble
Ali Log S	-7.27	-8.76	-8.45
Ali Solubility (mg/ml)	2.18E-05	1.00E-06	2.06E-06
Ali Solubility (mol/l)	5.36E-08	1.73E-09	3.57E-09
Ali Class	Poorly soluble	Poorly soluble	Poorly soluble
Silicos-IT LogSw	-8.54	-10.57	-10.56
Silicos-IT Solubility (mg/ml)	1.18E-06	1.57E-08	1.58E-08
Silicos-IT Solubility (mol/l)	2.90E-09	2.72E-11	2.74E-11
Silicos-IT class	Poorly soluble	Insoluble	Insoluble
GI absorption	Low	Low	Low
BBB permeant	No	No	No
Pgp substrate	No	No	No
CYP1A2 inhibitor	Yes	Yes	Yes
CYP2C19 inhibitor	Yes	Yes	Yes
CYP2C9 inhibitor	Yes	No	No
CYP2D6 inhibitor	Yes	No	No
CYP3A4 inhibitor	Yes	Yes	Yes
log Kp (cm/s)	-4.89	-5.4	-5.39
Lipinski #violations	1	1	1
Ghose #violations	1	3	3
Veber #violations	0	0	0
Egan #violations	1	1	1
Muegge #violations	1	1	1
Bioavailability Score	0.55	0.55	0.55
PAINS #alerts	0	0	0
Brenk #alerts	2	3	1
Leadlikeness #violations	2	2	2
Synthetic Accessibility	3.04	4.26	4.32

LIST OF PUBLICATION AND CONFERENCE PARTICIPATION

List of publication

Gemechu, S., Desalegn, T., Demissie, T. B., Ramachandran, V. P., & Eswaramoorthy, R. (2022). Metal Complexes in Target-Specific Anticancer Therapy: Recent Trends and Challenges. *Journal of Chemistry*, 2022, 1–19. <https://doi.org/https://doi.org/10.1155/2022/9261683>

Gemechu S., Demissie, T. B., Eswaramoorthy, R., Bogale, R. F., Kenasa, G., & Desalegn, T. (2023). *Biosynthesis of Silver Nanoparticles Functionalized with Histidine and Phenylalanine Amino Acids for Potential Antioxidant and Antibacterial Activities*. <https://doi.org/10.1021/acsomega.3c01910>

Gemechu S., Demissie, T. B., Koobotse, M., Kenasa, G., Beas, I. N., Zachariah, M., & Desalegn, T. (2024). Cytotoxic Cu(II) Complexes with a Novel Quinoline Derivative Ligand: Synthesis, Molecular Docking, and Biological Activity Analysis. *ACS Omega*, *Ii*. <https://doi.org/10.1021/acsomega.4c02129>

Shumi, G., Demissie, T. B., Kenasa, G., Lealem, A., Beas, I. N., & Desalegn, T. (2024). Synthesis, characterization, antibacterial, antioxidant and molecular docking studies of Zn(II) complexes with imine quinoline derivative ligand. *Inorganic Chemistry Communications*, *170*(2), 1–14. <https://doi.org/10.1016/j.inoche.2024.113304>

Contribution with other colleagues during my thesis work

Misganaw, K., Thillairasu, P., **Shumi, G.,** Debebe Habtamu, & Berhanu, S. (2024). Vanillin and 4-Nitroaniline Derived Schiff-Base and Its Nickel(II) Complex: Spectral Analysis and Antibacterial Investigation. *Bulletin of the Chemical Society of Ethiopia*, *38*(5), 1275–1289. <https://doi.org/https://dx.doi.org/10.4314/bcse.v38i5.7>

Takele, E., **Shumi, G.,** Mohammed, K., & Kenasa, G. (2024). Results in Chemistry Review on green synthesis , characterization , and antibacterial activity of CuO nanoparticles using biomolecules of plant extract. *Results in Chemistry*, *8*(June), 101606. <https://doi.org/10.1016/j.rechem.2024.101606>

Takele, E., Bogale, R. F., **Shumi, G.,** & Kenasa, G. (2023). Green Synthesis , Characterization , and Antibacterial Activity of CuO / ZnO Nanocomposite Using Zingiber officinale Rhizome Extract. *Journal of Chemistry*, 2023, 1–15. <https://doi.org/10.1155/2023/3481389>

List of conference participation

I have participating on two national and international conference to present the papers as following:



Paper presentation on The 1st African Biotechnology Congress on “Biotechnology for Sustainable Development of Africa” held June 6-7, 2024 at Elily International Hotel