REDOX-ACTIVE SILVER N-HETEROCYCLIC CARBENE COMPLEXES: A DUAL TARGETING ANTIBACTERIAL DRUG

A Thesis submitted in partial fulfillment of the requirements for the degree of Master of Science

by

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I HEREBY RECOMMEND THAT THE THESIS PREPARED UNDER MY
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ABSTRACT


Bacterial resistance to current β-lactam antibiotics necessitates new therapeutic approaches. Silver-based drugs offer a promising alternative because of their antibacterial properties and minimal bacterial resistance. However, many silver-based antibacterial drugs rapidly release silver ions, limiting the bioavailability of silver ions for a longer period of time. Recently, N-heterocyclic carbenes (NHC) containing silver complexes have become a popular subject of research due to their versatility and the slow release of silver ions under biological conditions. To further enhance the efficacy of silver-NHC drugs, we fused redox-active substituents such as naphthoquinone and ferrocenes to NHCs. Both naphthoquinone and ferrocenes have been shown to increase reactive oxygen species (ROS) under biological conditions that may augment the antibacterial action of redox-active silver-NHC complexes. Herein, we report synthesis and characterization of several redox-active silver-NHC complexes, 1-(ferrocenylmethyl)-3-mesityl-imidazol-2-ylidene silver chloride [(2)AgCl] (compound 6), 1,3-di-(ferrocenylmethyl)imidazol-2-ylidene silver chloride [(3)AgCl] (compound 7), bis (1-benzyl-3-mesitylimidazol-2-ylidene) silver chloride [(1)2Ag][Cl], (compound 9), Bis (1-(ferrocenylmethyl)-3-mesityl-imidazol-2-ylidene) silver chloride [(2)2Ag][Cl] (compound 10), Bis (1,3 di(ferrocenylmethyl)imidazole-2-ylidene) silver chloride [(3)2Ag][Cl]
(compound 11), and bis (1,3-dimesityl-4,5-naphthoquino-imidazol-2-ylidene) silver chloride [(4)₂Ag][Cl] (compound 12). Complexes were identified and confirmed via various analytical techniques, including ¹H NMR, ¹³C NMR, infrared spectroscopy, UV-vis spectroscopy, and single crystal X-ray crystallography.

A series of cyclic and differential pulse voltammetry experiments were conducted on the complexes to investigate their electrochemical properties. Complexes [(2)AgCl], [(3)AgCl], [(2)₂Ag][Cl], [(2)₂Ag][Cl], [(3)₂Ag][Cl], and [(4)₂Ag][Cl] exhibited redox activity attributed to the presence of ferrocene and naphthoquinone moiety. Their antibacterial properties were tested against gram-negative bacterial strains (Escherichia coli, Klebsiella pneumoniae, and Pseudomonas aeruginosa) and showed excellent antibacterial properties that are comparable to previously reported silver-NHC complexes. All silver-NHC complexes detailed in this report exhibit MIC and MBC values in the range of 2-12.5 µg/mL. Further mechanistic studies are necessary to elucidate the manner in which these complexes affect bacterial cells.
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Background

N-Heterocyclic Carbenes (NHCs)

N-heterocyclic carbenes (NHC) are cyclic compounds consisting of a carbene carbon and at least one nitrogen atom within the ring structure (Figure 1).\(^1,^2\) After the initial discovery, NHCs were often considered too reactive and unstable for isolation due to their incomplete electron octet and coordinative unsaturation at the carbon atom. Thus, multiple attempts to isolate and characterize NHCs dating back to 1830s were deemed unsuccessful.\(^3\) However, Wanzlick\(^4,^5\) revisited the field in the early 1960s, investigating the reactivity and stability of NHCs. This paved the way for synthesis of the first NHC transition metal complexes of chromium and mercury by Ofele\(^6\) and Wanzlick\(^7\) in 1968. The following decade saw minimal research in the area, however, and it was not until the successful isolation of the first stable crystalline NHC, 1,3-diamantylimidazole-2-yldene, by Arduengo\(^8\) in 1991 that the chemistry of NHCs began to receive widespread attention.

![Figure 1. General structure of NHC](image)

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\(^1\) Reference 1.
\(^2\) Reference 2.
\(^3\) Reference 3.
\(^4\) Reference 4.
\(^5\) Reference 5.
\(^6\) Reference 6.
\(^7\) Reference 7.
\(^8\) Reference 8.
Easy synthesis and exceptional stability of the first NHC sparked a wide range of experimental and theoretical studies, in which numerous NHCs were synthesized and investigated. While the compounds have since become popular in various disciplines, including polymer chemistry, coordination chemistry of transition metals, and optoelectronics, they have been used most commonly as ligands on transition metals for homogeneous catalysis. Recently, NHCs have proven useful in medicinal applications, as their versatility and stability allows them to serve as ligands on various transition metals for therapeutic applications.

The rise in the use of N-heterocyclic carbenes for a variety of applications stems from their stability and versatility. N-heterocyclic carbenes are strong σ-donating ligands, capable of binding strongly to transition metals. Moreover, the nitrogen atoms render this bond more stable than that of tri-alkyl/aryl phosphines. The electron-withdrawing nature of nitrogen, coupled with its p-electron donating property, stabilize the NHC both by lowering the energy of the s-orbital, and by donating electron density into the carbene’s empty p-orbital. Furthermore, NHCs were once considered only sigma donors, recent theoretical and structural analysis have revealed significant π-backbonding from the metal center to the C_carbene atom of the NHC, further contributing to its stability.

![Resonance structures of diamine NHCs](image)
In addition to their stability, NHC complexes also boast tremendous versatility, further contributing to their widespread use. This latter characteristic stems mainly from their structural diversity, as NHCs can be substituted with an array of functional groups at various positions, which in turn influence their lipophilicity,\textsuperscript{20} stability,\textsuperscript{21} and solubility of various complexes.\textsuperscript{22} For instance, kinetic stabilization can be achieved by the addition of steric and bulky substituents on the nitrogen atom, while electronic stabilization can be obtained by the addition of aromatic substituents to the backbone.\textsuperscript{23} Finally, redox activity can be achieved by the addition of redox-active components either to the nitrogen atom or the backbone. The ease with which NHCs can be fine-tuned to suit various uses is another reason that these compounds are becoming a staple molecule in chemistry.\textsuperscript{24}
Silver as an Antimicrobial

Silver has long served as an antimicrobial agent (Figure 4).\textsuperscript{25} Its use dates back to ancient times, when people stored their water in silver vessels to prevent microbial contamination.\textsuperscript{25,26} Around 700 A.C.E., it came into vogue as a treatment for burns and wounds.\textsuperscript{27} Much later, in the nineteenth century, doctors began using silver to prevent contagious diseases. Notable examples include Dr. Marion Sims, who introduced silver wires in surgery,\textsuperscript{28} and Dr. Carl Crede, a German gynecologist who began applying 2\% silver nitrate solution to eyes of newborns to prevent infection.\textsuperscript{29}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{figure4.png}
\caption{Medicinal uses of silver throughout history.}
\end{figure}

Although Alexander Fleming’s 1928 discovery of the first β-lactam antibiotic (Penicillin) revolutionized medicine, silver continued to be used as an antimicrobial. It was only in the 1940s, with the mass production and distribution of Penicillin to the
public, that the use of silver proved superfluous. Penicillin and its derivatives were highly effective at first, and Penicillin hailed worldwide as a miracle drug, able to treat many infectious diseases. Unfortunately, various cases of bacterial resistance soon arose. This alarming development spurred intense research into alternative antibacterial drugs. Silver ions reentered the medical field in the form of silver sulfadiazine (Figure 5) in 1968. Silver sulfadiazine, discovered by Dr. Charles Fox, is a silver-based antimicrobial agent that is effective against both gram-negative and gram-positive bacteria. It is commercially available as Silvadene® cream 1% and is used in hospitals worldwide to treat burns. It still remains the gold standard for silver-based antimicrobials. Silver has also been incorporated in wound dressings in nanocrystalline forms, a practice that helps sustain the release of silver to wound sites and increase the dressing’s antimicrobial power. Such dressings have shown to be effective in the treatment of leg ulcers and burn wounds.

![Silver Sulfadiazine Structure](image)

*Figure 5. Silver Sulfadiazine Structure.*

While reports of bacterial resistance towards silver have been rare, one study has shown unprecedent silver resistance by two strains of bacteria namely *Klebsiella pneumonia* and *Enterobacter cloacae*. These two organisms have shown to be capable of growth in the presence of high concentrations of silver ions. Further investigation of
this strain has revealed that it carries a plasmid pMG101 that is responsible for encoding two efflux pumps (SiICBA and SiIP). As a result, the strain is able to expel silver ions and prevent any subsequent damage to the bacterial cell.

An additional (and crucial) advantage of silver as an antimicrobial agent is that it is relatively non-toxic to humans in small doses; in fact, a small amount of silver (<2.43 mg/mL) can be found in an average human body. However, long and excessive exposure to elemental silver has been known to cause a rare cosmetic side effect. This condition is called argyria, consists of an irreversible gray-blue discoloration of the skin. In addition, toxicity studies have shown that high levels of silver in the circulatory system may affect the growth of connective tissue cells, bone marrow, and epidermal cells, by inhibiting cellular respiration.

Though silver’s widespread use as an antimicrobial agent dates back to the 20th century, silver ions’ mode of action is not yet clearly understood. However, various studies have attempted to shed light on the mechanism of action and many pathways have been proposed. For instance, silver ions may cause destruction of the peptidoglycan cell wall by interacting with the major proteins and enzymes responsible for cell wall synthesis. One study found that a loss of cell wall integrity was observed in yeast C. albicans when the silver cation interacted with the enzyme phosphomannose isomerase, an integral enzyme for cell wall synthesis. Furthermore, when silver cations enter the cell wall and the cytoplasm of bacteria, they have been shown to negatively affect metabolism and cell respiration, while also damaging DNA and thereby preventing bacterial replication. Yet another study revealed the effect of silver ions in the generation of reactive oxygen species (ROS). The study also demonstrated that silver...
ions are capable of generating superoxide radicals by damaging certain bacterial respiratory enzymes.\textsuperscript{44} The increase in ROS production can trigger a stress response and the activation of various apoptotic mechanism leading to bacteria death.\textsuperscript{44}

**Silver N-Heterocyclic Carbene Complexes**

The synthesis of the first silver NHC complex was reported in 1993 by Arduengo.\textsuperscript{45} The synthesis utilized a free carbene route (Scheme 1); however, due to the difficulty of generating free carbenes, as well as their sensitivity to various environmental factors, such as moisture and heat,\textsuperscript{46} other approaches were explored.\textsuperscript{47} This led to the most common method that is used for generation of silver-NHC complexes, the *in situ* deprotonation of imidazolium salt utilizing a basic silver precursor. Various bases have been used for this reaction, including silver carbonate and silver acetate,\textsuperscript{44} but silver oxide (Ag$_2$O) has been the most commonly used.\textsuperscript{47}

![Scheme 1. Synthesis of first silver NHC complex.](image)

The first usage of silver oxide as a base to synthesize silver NHC complexes is credited to Lin et al.,\textsuperscript{48} who synthesized 1,3 diethylbenzimidazole-2-ylidine (Scheme 2). The use of silver oxide in the synthesis of silver NHC complexes offers many advantages: the reaction is easier to conduct, it can be carried at ambient conditions, and
it can accommodate a variety of organic solvents.\textsuperscript{49} Thus, silver oxide is the preferred base for the generation of silver-NHC complexes.

![Scheme 2: Synthesis of first silver NHC complex using Ag\textsubscript{2}O as a base.]

The parameters that affect the aforementioned reaction include the equivalence of silver compared to the imidazolium salt, the source of silver used, solvent, and the reaction temperature.\textsuperscript{46} Various coordination numbers have been reported for silver-NHC complexes, two being the most common, followed by four, and rarely we encounter silver NHCs with coordination number of six.\textsuperscript{50}

The diversity of silver(I)-NHC structures and silver ion’s antibacterial properties facilitated the Young group for synthesis of the first antibacterial silver-NHC complexes in 2004 (Figure 6).\textsuperscript{51} These complexes were tested against various bacterial strains including \textit{Escherichia coli}, \textit{Pseudomonas aeruginosa}, and \textit{Staphylococcus aureus} and their minimum inhibitory concentration showed antibacterial properties comparable to silver nitrate MIC value range of 1-3 µg/mL.\textsuperscript{52}
This study led a surge of research and a library of silver-NHC complexes with antibacterial properties were synthesized and studied by various research groups.\textsuperscript{24} Taking advantage of NHCs’ versatility, complexes of varying structures and physical properties were synthesized, and their antimicrobial properties tested against various strains of bacteria.\textsuperscript{24}
Introduction

The use of silver as an antimicrobial agent can be traced to ancient times, when it was one of the few drugs capable of healing infections, burns, and wounds, as well as preventing contagious diseases. Silver in the form of silver nitrate was then used as an antiseptic in wound care for more than 200 years. Its use in medicine, however, diminished with the introduction of β-lactam antibiotics in the 1940s. While the mass production of penicillin and its derivatives was highly effective at first, bacterial resistance towards such antibiotics soon began to emerge. This development prompted a renewed interest in silver as an antimicrobial agent; it was reintroduced into the field with Fox’s 1968 discovery of silver sulfadiazine (Silvadene®).

Silver sulfadiazine is a silver antimicrobial agent that is currently considered the gold standard for silver-based antimicrobials, used in hospitals worldwide to treat burns. Moreover, bacterial resistance to silver is extremely rare, and the substance poses no threat to humans (except for a very uncommon skin condition called argyria, which only occurs after prolonged and excessive exposure). Despite their advantages, the use of silver nitrate, silver sulfadiazine, and other related silver compounds were limited as they are exclusively targeted for external applications due to the fact that they are water insoluble.
However, recent research has focused on the discovery of new and more effective silver-based antibacterial compounds and, in 2004, Young’s group reported silver(I) N-heterocyclic carbene complexes as a new class of antibiotics. N-heterocyclic carbenes (NHC) form strong M–C_{carbene} bonds that are far more stable than most phosphines due to their increased σ-donation, as well as π-back-donation from metal to the carbene carbon.\textsuperscript{58,59,60} This allows for a slower release of silver ions, resulting in an increased exposure time that disrupts cell wall synthesis and prevents bacterial growth.\textsuperscript{61} This difference renders silver(I)-NHC complexes more effective against bacterial reinfection than traditional silver-based compounds. Moreover, NHCs can be easily modified to accommodate various substituents, such as caffeine\textsuperscript{62} and chlorine\textsuperscript{63} amongst many others, allowing for further modification of bioactivity and bioavailability. For instance, the electron-withdrawing chlorine groups have been shown to increase the stability of silver-NHC complexes in a water medium from a couple of hours to 17 weeks,\textsuperscript{63} while the use of caffeine (a xanthine derivative) as the NHC backbone generates a biocompatible complex, since xanthines are used medicinally and have low toxicity to humans.\textsuperscript{62} These properties suggest that silver(I)-NHCs may hold great promise in the field of antibacterial research.

The N-heterocyclic carbene was first reported in the 1960s by Öfele\textsuperscript{6} and Wanzlick\textsuperscript{7}, but it was only in the early 1990s, after the isolation of the first stable carbene by Arduengo,\textsuperscript{45} that its use became more common. This discovery led to important developments in the coordination chemistry of NHCs.\textsuperscript{11} Numerous metal-NHC complexes have since been prepared and used as homogeneous catalysts\textsuperscript{13} in organometallic transformations\textsuperscript{13} and, more recently, the use of NHC ligands has become
prevalent in biological applications.\textsuperscript{64} The stability and versatility of these ligands allows them to serve as metal carriers for transition metals such as copper, gold, and silver in biological media.\textsuperscript{14}

Building on this foundation, we synthesized several silver-NHC complexes fused with ferrocene and naphthoquinone substituents. Ferrocene is an organometallic compound that is capable of undergoing one-electron oxidation to form a ferrocenium cation, which then goes through a one-electron reduction to regenerate ferrocene.\textsuperscript{65} Similarly, naphthoquinone, a redox-active moiety that is characterized by the presence of two carbonyl groups, exhibits multiple reversible redox-active capabilities.\textsuperscript{66}

The reversible redox-active properties of ferrocene and naphthoquinone in biological media have proven effective against various diseases, including cancer.\textsuperscript{66} The redox activity increases reactive oxygen species (ROS), causing oxidative stress in cells and leading to cell proliferation inhibition and even cell death.\textsuperscript{66,67,68} The accumulation of highly reactive ROS such as hydrogen peroxide (H$_2$O$_2$) and O$_2^−$ leads to oxidative damage on biomolecules such as proteins, lipids, and nucleic acid and further promotes the activation of programmed cell death (PCD).\textsuperscript{69}
We hypothesized that an NHC complex that makes use of both redox-active substituents and the antimicrobial properties of silver could synergistically inhibit bacterial growth (Figure 7). To this end, a dual targeting drug with the ability to inhibit bacterial growth through multiple pathways is an appealing prospect in drug discovery, one that could open the door to future antibacterial drugs.
Experimental Section

General Considerations: All synthetic procedures were executed under a nitrogen atmosphere glove box (Inert Glove Box System). All glasswares were subjected to heat at 110 °C for 12 h before use. The compounds: 1,3-dimesitylnapthoquinimidazolium chloride, 4, [4H][Cl], 70 1-(benzyl)-3-(2,4,6-trimethylphenyl) imidazolium chloride, 71 1,[1H][Cl] 1,3-di(ferrocenylmethyl)imidazolium iodide, 72 and 1-(ferrocenylmethyl)-3-mesityl-limidazolium iodide 73 were prepared according to the literature procedures. Solvents (CH$_2$Cl$_2$, Et$_2$O, THF, and toluene) were dried with a solvent purification system (Inert Innovative Technology, Inc), degassed using three consecutive freeze-pump-thaw cycles and stored over 4 Å molecular sieves in the glove box. The NMR solvents: CDCl$_3$ (99.9%) and MeOD (99.8%) were purchased from Acros Laboratories, dried over 4 Å molecular sieves and stored in the glove box prior to use. All other chemicals were purchased commercially and used as received. UV-vis spectra were achieved at room temperature with a Varian Cary 50 Bio UV-vis spectrophotometer. Molar absorptivity for compounds [(2)AgCl], [(3)AgCl], [(2)2Ag][Cl], [(3)2Ag][Cl], and [(4)2Ag][Cl] were reported in M$^{-1}$ cm$^{-1}$. $^1$H and $^{13}$C NMR spectra were recorded on Bruker 300 MHz spectrometer. Spectra were referenced to the residual solvent as an internal standard, for $^1$H NMR: CDCl$_3$, 7.26 ppm and $^{13}$C NMR 77.16 ppm and for $^1$H NMR: MeOD, 3.31 ppm and $^{13}$C NMR 49.15 ppm. Electroanalytical assessments were performed using a
CHI620E electrochemical workstation using a silver wire quasi-reference electrode, a platinum disk working electrode and a Pt wire auxiliary electrode in a gas tight three-electrode cell under a nitrogen atmosphere. The measurements were performed using 1.0 mM of the analyte in dry DMSO with 0.1 M [N(nBu)₄][PF₆] as the electrolyte and decamethylferrocene (Fc*) as the internal standard for complexes [(2)AgCl], [(3)AgCl], [(2)₂Ag][Cl], [(3)₂Ag][Cl], and ferrocene (Fc) as the internal standard for complex [(4)₂Ag][Cl]. Differential pulse voltammetry measurements were performed with 100 mV pulse amplitudes and 2 mV data intervals. All potentials recorded herein were calculated by differential pulse voltammetry at 100 mVs⁻¹ scan rates and referenced to a saturated calomel electrode (SCE) by shifting decamethylferrocene ⁰⁺ to −0.030 (DMSO) and by shifting ferrocene ⁰⁺ to 0.435 V (DMSO).⁷⁴

**Synthesis of Imidazolium Chloride Salts**

1-(Ferrocenylmethyl)-3-mesityl-imidazolium chloride, (2)[H][Cl], 2. To an Erlenmeyer flask equipped with stir bar and 75 mL MeOH, 1-(ferrocenylmethyl)-3-mesityl-imidazolium iodide (0.780 g, 1.52 mmol) was added with amberlite IRA-400 chloride ion exchange resin (7.80 g). The resulting mixture was stirred for 1 h at room temperature. The resin was filtered and washed with methanol and the combined filtrates were charged with a fresh resin (7.80 g) and stirred for 1 h at room temperature. This process was repeated two times. The final filtrate was dried under reduced pressure. The dried yellow residue was dissolved in minimum amount of CH₂Cl₂ and product was precipitated with 15 mL Et₂O producing a yellow solid. Yield: 0.621 g, 97 %. ¹H NMR (δ, CDCl₃, 300 MHz): δ 10.97 (s, 1H), 7.50 (s, 1H), 7.03 (s, 1H), 6.95 (s, 2H), 5.57 (s,
2H), 4.51 (s, 2H), 4.27-4.24 (d, 7H), 2.30 (s, 3H), 2.02 (s, 6H). $^{13}$C NMR (δ, CDCl$_3$, 75 MHz): δ 141.18, 138.34, 134.23, 130.90, 129.88, 122.83, 121.98, 79.42, 69.71, 69.40, 50.15, 21.12, 17.81.

1,3-di (ferrocenylmethyl)imidazolium chloride, (3)[H][Cl], 3. To an Erlenmeyer flask equipped with stir bar and 75 mL methanol, 1, 3-di (ferrocenylmethyl)imidazolium iodide (0.550 g, 0.928 mmol) was added with amberlite IRA-400 chloride ion exchange resin (5.50 g). The resulting mixture was allowed to stir for 1 h at room temperature. The resin was filtered and washed with methanol and the combined filtrates were charged with a fresh resin (5.50 g) and stirred for 1 h at room temperature. This process was repeated two times. The final filtrate was dried under reduced pressure. The dried yellow residue was dissolved in minimum amount of CH$_2$Cl$_2$ and product was precipitated with 15 mL Et$_2$O producing a yellow solid. Yield: 0.441 g, 95 %. $^1$H NMR (δ, CDCl$_3$, 300 MHz): δ 11.23 (s, 1H), 6.89 (s, 2H), 5.29 (s, 4H), 4.28 (s, 4H), 4.23 (s, 14H). $^{13}$C NMR (δ, CDCl$_3$, 75 MHz): δ 120.38, 78.81, 69.88, 69.71, 69.37, 50.18.

Synthesis of Heterolyptic Imidazole-2-Ylidene Silver Chlorides$^{70}$

1-(ferrocenylmethyl)-3-mesityl-imidazol-2-ylidene silver chloride, [(2)AgCl], 6. A 10 mL vial with stir bar was charged with 1-(ferrocenylmethyl)-3-mesityl-imidazolium chloride (0.150 g, 0.356 mmol, 1 eq), silver oxide (0.061 g, 0.267 mmol, 0.75 eq), and 4.0 mL of dry CH$_2$Cl$_2$. The mixture was stirred at room temperature for 24 h, which resulted in a solution with yellow suspension. The heterogeneous mixture was filtered through a plug of celite into a pre-weighed vial and the filtrate was concentrated to a minimum amount (~ 2 mL) and the compound was precipitated with 15 mL of Et$_2$O. The
precipitate was washed with $3 \times 10 \text{ mL}$ of Et$_2$O to yield a yellow solid. Yield: 0.125 g, 89 %. $^1$H NMR ($\delta$, CDCl$_3$, 300 MHz): $\delta$ 7.08 (s, 1H), 6.93 (s, 2H), 6.85 (s, 1H), 5.15 (s, 2H), 4.33 (s, 2H), 4.25-4.23 (m, 7H), 2.32 (s, 3H), 1.93 (s, 6H).$^{13}$C NMR ($\delta$, CDCl$_3$, 75 MHz): $\delta$ 139.68, 135.54, 134.82, 129.58, 122.67, 122.57, 120.45, 81.57, 69.34, 69.16, 52.14, 21.19, 17.84.

1,3-di(ferrocenylmethyl)imidazol-2-ylidene silver chloride, [(3)AgCl], 7. A 10 mL vial with stir bar was charged with 1,3-di(ferrocenylmethyl)imidazolium chloride (0.145 g, 0.289 mmol, 1 eq), silver oxide (0.050 g, 0.217 mmol, 0.75 eq), and 4.0 mL of dry CH$_2$Cl$_2$. The mixture was stirred at room temperature for 24 h, which resulted in a solution with yellow suspension. The heterogeneous mixture was filtered through a plug of celite into a pre-weighed vial and the filtrate was concentrated to a minimum amount (~ 2 mL) and the compound was precipitated with 15 mL of Et$_2$O. The precipitate was washed with $3 \times 10 \text{ mL}$ of Et$_2$O to yield a yellow solid. Yield: 0.112 g, 85 %. $^1$H NMR ($\delta$, CDCl$_3$, 300 MHz): $\delta$ 6.81 (s, 2H), 4.96 (s, 4H), 4.28-4.27 (m, 4H), 4.20-4.19 (m, 14H).$^{13}$C NMR ($\delta$, CDCl$_3$, 75 MHz): $\delta$ 120.30, 81.55, 69.30, 69.27, 69.09, 52.07.

Synthesis of Homoleptic Imidazole-2-Ylidene Silver Chlorides

Bis(1-benzyl-3-mesitylimidazol-2-ylidene) silver chloride, [(1)$_2$Ag][Cl], 9. In a 10 mL vial equipped with a stir bar, 1-(benzyl)-3-(2,4,6-trimethylphenyl) imidazolium chloride (0.120 g, 0.384 mmol, 1 eq) and Sodium bis(trimethylsilyl)amide (0.077 g, 0.422 mmol, 1.1 eq) were mixed in toluene. After 2 h, the yellow solution was filtered through a plug of celite into a vial containing AgCl (0.0247 g, 0.173 mmol, 0.45 eq). The mixture was stirred for 24 h. The resulting solution was filtered through a plug of celite and dried under vacuum. The brown residue was dissolved in minimum amount (~ 2 mL) of
CH₂Cl₂ and the product was precipitated with 15 mL Et₂O and further washed with 3 × 10 mL Et₂O to produce a white solid. Yield: 0.75 g, 78%. ¹H NMR (δ, CDCl₃, 300 MHz): δ 7.47 (m, 2H), 7.30-7.27 (m, 6H), 7.13-7.12 (m, 4H), 6.88 (s, 2H), 6.84 (s, 4H), 5.28 (s, 4H), 2.29 (s, 6H), 1.77 (s, 12H).¹³C NMR (δ, CDCl₃, 75 MHz): δ 139.29, 136.71, 135.75, 134.98, 129.27, 129.02, 128.39, 127.62, 122.76, 55.26, 21.23, 17.73.

Bis (1-(ferrocenylmethyl)-3-mesityl-imidazol-2-ylidene) silver chloride, [(2)₂Ag][Cl], 10. In a 10 mL vial equipped with a stir bar was added 1-(ferrocenylmethyl)-3-mesitylimidazolium chloride (0.142 g, 0.337 mmol, 1 eq), sodium bis(trimethylsilyl)amide (0.0681 g, 0.371 mmol, 1.1 eq), and 5 mL of toluene. After 2 h stirred at rt, the yellow solution was filtered through a plug of celite into a vial containing AgCl (0.0217 g, 0.152 mmol, 0.45 eq). The mixture was stirred for 24 h. The resulting solution was filtered through a plug of celite and dried under vacuum. The brown residue was dissolved in minimum amount (~ 2 mL) of CH₂Cl₂ and the product was precipitated with 15 mL Et₂O and further washed with 3 × 10 mL Et₂O to produce a yellow solid. Yield: 0.96 g, 82%. ¹H NMR (δ, CDCl₃, 300 MHz): δ 7.55-7.54 (m, 2H), 6.89 (s, 4H), 6.89-6.84 (m, 2H), 5.13 (s, 4H), 4.21-4.16 (m, 18H), 2.36 (s, 6H), 1.77 (s, 12H).¹³C NMR (δ, CDCl₃, 75 MHz): δ 139.20, 135.87, 134.99, 129.58, 129.32, 122.48, 122.41, 122.31, 83.02, 69.18, 68.92, 68.67, 51.49, 21.33, 17.78.

Bis (1,3 di (ferrocenylmethyl)imidazole-2-ylidene) silver chloride, [(3)₂Ag][Cl], 11. In a 10 mL vile equipped with a stir bar, 1,3 di (ferrocenylmethyl)imidazolium chloride (0.200 g, 0.400 mmol, 1 eq) and Sodium bis(trimethylsilyl)amide (0.0806 g, 0.440 mmol, 1.1 eq) were mixed in toluene. After 2 h, the yellow solution was filtered through a plug
of celite into a vile containing AgCl (0.0257 g, 0.180 mmol, 0.45 eq). The mixture was stirred for 24 h. The resulting solution was filtered through a plug of celite and dried under vacuum. The brown residue was dissolved in minimum amount (~ 2 mL) of CH$_2$Cl$_2$ and the product was precipitated with 15 mL Et$_2$O and further washed with 3 × 10 mL Et$_2$O to produce a yellow solid. Yield: 0.120 g, 60%. $^1$H NMR (δ, CDCl$_3$, 300 MHz): δ 6.83 (s, 4H), 4.99 (s, 8H), 4.28–4.27 (m, 8H), 4.20–4.19 (28H). $^{13}$C NMR (δ, CDCl$_3$, 75 MHz): δ 120.32, 81.63, 69.27, 69.10, 52.04.

**Bis (1,3-dimesityl-4,5-naphthoquinon-imidazol-2-ylidene) silver chloride, [(4)$_2$Ag][Cl], 12.** In a 10 mL vile equipped with a stir bar, 1,3-dimesitylnaphthoquinimidazolium chloride (0.150 g, 0.318 mmol, 1 eq) and Sodium bis(trimethylsilyl)amide (0.064 g, 0.349 mmol, 1.1 eq) were mixed in toluene. After 24 h, the yellow solution was filtered through a plug of celite into a vile containing AgCl (0.020 g, 0.14 mmol, 0.45 eq). The mixture was stirred for 24 h. The resulting solution was filtered through a plug of celite and dried under vacuum. The brown residue was washed with 10 mL of toluene followed by 3 × 10 mL Et$_2$O. The solid was dissolved in minimum MeOH, filtered through a celite and dried under vacuum to produce a green solid. Yield: 0.09 g, 64%. $^1$H NMR (δ, CD$_3$OD, 300 MHz): δ 8.08–8.06 (m, 4H), 7.84–7.81 (m, 4H), 7.10 (s, 8H), 2.50 (s, 12H), 1.77 (s, 24H).$^{13}$C NMR (δ, CD$_3$OD, 75 MHz): δ 175.92, 141.69, 135.93, 135.63, 135.00, 134.23, 134.15, 133.77, 130.68, 128.05, 21.53, 17.87. IR (cm$^{-1}$): 1683 (MeOH).

**In Vitro Antimicrobial Activity**

Using an agar dilution procedure,$^{75}$ the antimicrobial activity of the new silver-NHC complexes were investigated. The minimal inhibitory concentrations (MICs) and the
minimal bactericidal concentrations (MBCs) for complexes 1-12 were tested against standard bacterial strains (E. Coli, P. aeruginosa, K. pneumoniae). Bacteria from fresh overnight plates were suspended in standard Muller-Hinton broth (M-H) to an optical density at 600 nm (OD_{600}) of 0.13. The bacteria were diluted in the broth to a concentration of 10^2 CFU/mL. The stock solutions of all complexes were prepared in DMSO (25%). The concentrations of the tested complexes were 1, 2, 4, 5, 8, 10, 12.5, 25, 50, 60, 70, 80, 90 and 100 µg/mL. The MIC was the lowest concentrations, at which visible growth is absent after incubation of the plate for 24 h at 37 °C. The MBC for all complexes were determined by plating all of the clear wells of the MIC experiments on TSA agar plates, incubating the plates overnight at 37 °C, and observing the concentration at which there were no visible colony growth. Silver nitrate was used as the control. A total of 3 trials were conducted for each experiment.
Results and Discussion

Figure 8. Silver-NHC complexes studied.

Synthesis and Characterization

Using a modified literature procedure\textsuperscript{70}, a neutrally charged silver-NHC complex [(2)AgCl] (6) and [(3)AgCl] (7) were synthesized (Scheme 3).
Complex [(2)AgCl] (6) and [(3)AgCl] (7) were prepared by treating 1 equivalence of compound (2)[H][Cl] and (3)[H][Cl], respectively, with 0.75 Ag₂O and molecular sieves for 24 h at room temperature in CH₂Cl₂. At the end of 24 h stir, the crude mixture was subjected to simple filtration to yield crude solids of [(2)AgCl] (6) and [(3)AgCl] (7). The obtained crude products were dissolved in a minimum amount of CH₂Cl₂ and precipitated with Et₂O to yield microcrystals of [(2)AgCl] (6) and [(3)AgCl] (7). Proton NMR spectral analyses of complex [(2)AgCl] (6) and [(3)AgCl] (7) in CDCl₃ proved consistent with the molecular structure. Specifically, the absence of the hydrogen attached to C_carbene in the ¹H NMR of [(2)AgCl] (6) and [(3)AgCl] (7), corroborates the formation of silver-carbene bond at the carbene carbon. ¹H NMR analysis of complex [(2)AgCl] (6) displayed mesityl hydrogens (ortho-CH₃ and para-CH₃) at 1.93 and 2.32 ppm, respectively. The signals between 4.23-4.33 ppm represents the 9 hydrogens attached to the ferrocene unit. The benzylic CH₂ hydrogens were observed at 5.15 ppm. The signals at 6.85 and 7.08 ppm each correspond to C2 and C3 imidazole hydrogens. The two aromatic mesityl hydrogens, are represented by a signal at 6.93 ppm. Proton NMR spectrum of compound [(3)AgCl] (7) reveals signals between 4.19-4.28 ppm which
corresponds to 18 hydrogens attached to the two ferrocene units. The symmetrical benzylic CH₂ hydrogens is represented by a single peak at 4.96 ppm. The two hydrogens at C2 and C3 of the imidazole unit is represented by a single peak at 6.81 ppm, consistent with C2 symmetry is present due to the two ferrocene substituents, in contrast to complex [(2)AgCl] (6), where the hydrogens at C2 and C3 is presented by two peaks validating the asymmetric nature of the complex.

![Image](image.png)

*Figure 9. ¹H NMR spectrum (300 MHz, CDCl₃) of compound 6, [(2)AgCl].*

The homoleptic complexes 9-12 were synthesized using a modified literature procedure by *in situ* generation of free carbene of the corresponding imidazolium salts 1-4 using NaN(SiMe₃)₂, followed by immediate treatment with 0.40 equivalents of AgCl in toluene (Scheme 4).
The insoluble nature of complexes 9-12 in toluene allowed for their separation from the reaction mixture. The complexes were subjected to celite filtration followed by 3 × 10 mL toluene and 3 × 10 mL Et₂O washes. Subsequently, complexes 9-11 were dissolved in minimum amount (~ 2 mL) of CH₂Cl₂ and precipitated with 10 mL Et₂O. While, complex [(4)₂Ag][Cl] (12) was dissolved in MeOH, filtered using celite plug, and dried. 

$^1$H NMR spectral analyses of the homoleptic complexes proved consistent with molecular structures. The absence of the hydrogen attached to $C_{\text{carbene}}$ in the $^1$H NMR of complexes 9–12 proves the formation of silver-carbene bond at the carbene carbon. Complex [(1)₂Ag][Cl] (9) proton NMR (CDCl₃) spectrum shows mesityl hydrogens ($ortho$-CH₃ and $para$-CH₃) were observed at 1.77 and 2.36 ppm respectively. The signals between 4.16-4.21 ppm represents the 9 hydrogens attached to the ferrocene unit. The benzylic
CH₂ hydrogens was observed at 5.13 ppm. The signals at 6.85 and 7.54 ppm each correspond to C2 and C3 imidazole hydrogens. The two aromatic mesityl hydrogens, are represented by a signal at 6.89 ppm. Complex [(2)₂Ag][Cl] (10) ¹H NMR (CDCl₃) spectrum reveals signals between 4.19-4.28 ppm which corresponds to the 18 hydrogens attached to the two ferrocene units. The symmetrical benzylic CH₂ hydrogens is represented by a single peak at 4.99 ppm. The two hydrogens at C2 and C3 of the imidazole unit is represented by a single peak at 6.83 ppm proving a symmetry is present due to the two ferrocene substituents. ¹H NMR (CDCl₃) spectrum for complex [(3)₂Ag][Cl] (11) shows mesityl hydrogens (ortho-CH₃ and para-CH₃) were observed at 1.77 and 2.29 ppm respectively. The benzylic CH₂ hydrogens was observed at 5.28 ppm. The mesityl aromatic hydrogens are represented at 6.84 ppm. Imidazolylidene hydrogens C2 and C3 appear at 6.88 and 7.47 ppm respectively. The hydrogens of four phenyl groups are represented in the region 7.10 – 7.30 ppm. Proton NMR spectral analyses of complex [(4)₂Ag][Cl] (12) in MeOD proved consistent with the molecular structure. For instance, mesityl hydrogens (ortho-CH₃) were observed at 1.77 ppm, while mesityl hydrogens (para-CH₃) were observed at 2.50 ppm. The mesityl aromatic hydrogens appeared at 7.10 ppm, while the naphthoquinone benzene hydrogens appeared within the 7.81-8.09 ppm range. To further confirm the validity of the structure, an IR spectrum of complex [(4)₂Ag][Cl] (12) was obtained in MeOH. The IR spectrum revealed a peak at 1683 cm⁻¹, which is indicative of a carbonyl peak from the naphthoquinone moiety. This peak is consistent with literature⁷⁰ and provides additional validation of the complex synthesized.
The absence of C–Ag in the $^{13}$C NMR in all the complexes is indicative of fluxional behavior which is a common phenomenon that is observed with many silver (I) complexes.\textsuperscript{77}

![Figure 10. $^{13}$C NMR spectrum (75 MHz, CDCl$_3$) of compound 6, [(2)AgCl].](image)

To further substantiate the validity of the complexes synthesized, x-ray diffractions of the heterolytic complexes 6-7 and the homoleptic complexes 9-12 were obtained. For instance, single crystals of complex [(2)AgCl] (6) were grown by slowly diffusing hexane into a concentrated CHCl$_3$ solution. Complex [(2)AgCl] (6) has an C1–Ag1–Cl1 angle of 175.84(7) $^\circ$ and N2–C1–N1 angle of 104.4 (2) $^\circ$. The C1–N1 and C2–N1 bond lengths are 1.355 (3) Å and 1.386 (4) Å, respectively. The Bond length for C1–Ag1 is 2.064 (3) Å and bond length for Ag1–Cl1 is 2.320 (7) Å (Figure 9). The Ag-C bond
distance of 2.064(3) Å is comparable to those found in other heterolytic silver NHC complexes.\textsuperscript{78}

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{complex6_ortep.png}
\caption{ORTEP diagram of complex 6 drawn using POV-Ray. Thermal ellipsoid plots are drawn at 50\% probability level and hydrogen atoms are omitted for clarity.}
\end{figure}

Similarly, single crystals of [(1)\textsubscript{2}Ag][Cl] (9) were grown by diffusing methyl \textit{tert}-butyl ether into a concentrated CHCl\textsubscript{3} solution. Complex 9 has an N1–C1–N2 bond angle of 104.09 (19) Å, C1–Ag1–Cl1 bond angle of 101.06 (6) °, and Ag1–Cl1–Ag2 bond angle of 148.87 (2) °. The bond lengths between C1–Ag1 is 2.104 (2) Å, Ag1–Cl1 is 2.8152 (6) Å, Cl1–N1is 1.350 (3) Å, Cl1–Ag2 is 2.8754 (6) Å, and N1–C2 is 1.382 (3) Å (Figure 10).
Figure 12. ORTEP diagram of complex 9 drawn using POV-Ray. Thermal ellipsoid plots are drawn at 50% probability level and hydrogen atoms are omitted for clarity.

Single crystals for Complex [(2)2Ag][Cl] (10) were grown by diffusing methyl tert-butyl ether into a concentrated CHCl3 solution. Complex [(2)2Ag][Cl] (10) presented a bond angle of 165.99 (17)° for C1–Ag1–C4 and a bond angle of 104.3 (4)° for N1–C1–N2. The bond lengths between Ag1–C1 is 2.089 (4) Å, C1–N1 is 1.346 (6) Å, N1–C2 is 1.389 (6) Å, and C2–C3 is 1.343 (7) Å (Figure 11).
Silver-NHC complexes \([\text{(2)AgCl}] \) (6), \([\text{(3)AgCl}] \) (7), \([\text{(2)\text{Ag}[Cl]}] \) (10), \([\text{(3)\text{Ag}[Cl]}] \) (11), and \([\text{(4)\text{Ag}[Cl]}] \) (12) were also characterized by ultraviolet-visible spectroscopy. Complexes 6-7 and 10-11, consisting of ferrocene moiety, displayed an absorption band at ~ 440 nm. This is consistent with values reported in the literature for ferrocene containing complexes.\(^6\) Complex 10, which contains naphthoquinone moiety, displayed a low energy \(\pi\) to \(\pi^*\) band near the visible region at ~ 340 nm and a high energy \(\pi\) to \(\pi^*\) band at ~ 260 nm. These peaks corresponding to naphthoquinone moiety are consistent with literature.\(^7\)
**Electrochemistry**

To analyze the electronic properties of the complexes synthesized, a series of electrochemical analysis, including cyclic voltammetry (CV) and differential pulse voltammetry (DPV) were carried out with \([N(nBu)_4][PF6]\) in anhydrous dimethyl sulfoxide (DMSO). Cyclic voltammetry of the redox-active complexes synthesized are provided in **Figure 12**, while key data obtained from differential pulse voltammetry for the complexes are summarized in **Table 1**. The heterolytic complexes 6-7 and the homoleptic complex 10-11 that includes a ferrocene substituent, displayed a reversible one electron oxidation \(\text{Fe}^{2+} \rightarrow \text{Fe}^{3+}\). One electron oxidation for complex \([(2)\text{AgCl}] (6)\) and \([(3)\text{AgCl}] (7)\) were observed at \(\sim 0.46 \text{ V} \) (vs. SCE), whereas the homoleptic complexes \([(2)_2\text{Ag}[\text{Cl}] (10)\) and \([(3)_2\text{Ag}[\text{Cl}] (11)\), their oxidations were observed at 0.56 and 0.52 V (vs. SCE) respectively.
Figure 14. Cyclic voltammetry of redox-active complexes
Table 1. Electrochemical properties.

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<th>COMPOUND</th>
<th>E_{1/2} (V)</th>
<th>DPV</th>
</tr>
</thead>
<tbody>
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<td></td>
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<tr>
<td>[(3)AgCl], 7</td>
<td>0.46</td>
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<td></td>
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<tr>
<td>[(4)\textsubscript{2}Ag][Cl], 12</td>
<td>$-0.48^a$, $-1.34^b$</td>
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</tr>
</tbody>
</table>

*a* ASSIGNED AS THE FIRST REDUCTION (FORMATION OF SEMIQUINONE RADICAL, NQ$^-$)

*b* ASSIGNED AS THE SECOND REDUCTION (FORMATION OF QUINONE DIANION, NQ$^{2-}$)

Electrochemical analysis of the redox-active complexes. The potentials were obtained from differential pulse voltammetry measurement in DMSO using 0.1 M [N(nBu)$_4$]$^+$[PF$_6$]$^-$ as the supporting electrolyte, 0.1 mM analyte, and referenced vs. SCE. For the corresponding cyclic voltammograms and differential pulse voltammograms refer to the supporting information.

Compound [(4)\textsubscript{2}Ag][Cl] (12) exhibited cathodic waves that occur in two sequential steps. The first wave is completely reversible, and the second wave is quasireversible at 0.1 mV s$^{-1}$ scan rate. These electrochemical features are attributed to the reduction of the quinone moiety to first produce the semi-quinone radical (NQ$^-$) at −0.48 V and then produce the quinone dianion (NQ$^{2-}$) at −1.34 V.

The electrochemical properties of the complexes [(2)AgCl] (6), [(3)AgCl] (7), [(2)\textsubscript{2}Ag][Cl] (10), [(3)\textsubscript{2}Ag][Cl] (11), and [(4)\textsubscript{2}Ag][Cl] (12) from the series of CV and DPV conducted, confirms the redox-active potential of the ferrocene and naphthoquinone substituents that are present in these complexes.
Antibacterial Activity

After the synthesized complexes were confirmed via analytical techniques and their redox-activity studied, the in vitro antibacterial properties of these complexes were investigated. In order to determine the effectiveness of these heterolytic and homoleptic complexes, minimum inhibitory concentration (MIC) and minimum bacteriostatic concentrations (MBC) were determined against three gram-negative bacteria (E. coli, K. pneumoniae, P. aeruginosa) in Muller-Hinton (M-H) broth (Table 2). Complexes were dissolved in DMSO and a series of dilutions were prepared followed by 24 h incubation with the bacterial strain in growth media at 37 °C. The MIC was determined as the lowest concentrations, at which visible growth is absent. The MBC for all complexes were determined by plating all of the clear wells of the MIC experiments on TSA agar plates, incubating the plates overnight at 37 °C, and observing the concentration at which there were no visible colony growth.

The imidazolium salts 1-4, containing no silver ions, had MIC and MBC values ranging from 60-100±5 µg/mL. These salts exhibited high MIC and MBC values due to the absence of the antibacterial silver ions. The heterolytic complexes 5-8 and the homoleptic complexes 9-12 displayed MIC and MBC values more effective than their corresponding salts and comparable to silver nitrate—an established antibacterial agent. The MIC value range for complexes 5-8 was 4-8 µg/mL with the most effective complexes being complex [(2)AgCl] (6), [(3)AgCl] (7) across all three bacteria. These complexes also proved to be bactericidal against the bacteria tested with MBC values range from 2-8 µg/mL. Similarly, the homoleptic complexes 9-12 exhibited antibacterial properties superior than their corresponding imidazolium salts and comparable to the
heterolytic complexes and silver nitrate. Complexes 9-12 displayed antibacterial properties with MIC and MBC range of 2-12.5±2 µg/mL.

Overall, these MIC and MBC studies suggests that these complexes appear to be capable of killing gram-negative bacteria at concentrations comparable to silver NHCs values obtained the literature.52 For instance, Young’s silver NHCs have MIC values ranging from 1-8 µg/mL.51 We propose that both the redox-active substituents and silver ions have contributed to the bacterial growth inhibition. In order to further elucidate on the mechanism, mechanistic studies and evaluations need to be conducted.
Table 2. MIC and MBC of the complexes studied in µg/mL.

<table>
<thead>
<tr>
<th>Species</th>
<th>E. coli</th>
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<th>P. aeruginosa</th>
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<td>MBC</td>
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<td>AgNO₃</td>
<td>2±2</td>
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</tr>
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</table>
Conclusion

We report the synthesis of six redox-active silver NHC complexes with antibacterial properties. The heterolytic complexes, [(2)AgCl] and [(3)AgCl], were prepared by the reaction of their respective imidazolium salts with silver (I) oxide in dichloromethane; while the homoleptic complexes, [(1)2Ag][Cl], [(2)2Ag][Cl], [(3)2Ag][Cl], and [(4)2Ag][Cl], were synthesized by a two-step process, which included the deprotonation of salts followed by silver chloride treatment. All complexes were confirmed and characterized using elemental analysis and spectroscopic techniques including, 1H and 13C NMR spectroscopy, infrared spectroscopy, and UV-vis Spectroscopy. 1H and 13C NMR spectral analyses of the heterolytic and homoleptic complexes proved consistent with molecular structures. The electronic properties of the complexes were investigated by cyclic and differential pulse voltammetry. Complexes with naphthoquinone or ferrocene substituents exhibited redox-active properties consistent with literature. The antibacterial properties of these complexes were tested against three standard strains of gram-negative bacteria (E. coli, K. pneumoniae, P. aeruginosa). All complexes exhibited similar MIC and MBC activities to silver nitrate, establishing the antibacterial properties of these complexes.

Our future work involves examining and understanding the mechanistic pathway these redox-active silver-NHC complexes employ to inflict bacterial death. Further, since
redox-active complexes have shown to cause apoptosis to cancer cells, our research group is interested in investigating the anticancer properties of these complexes.
Analytical Data

Electrochemical Analysis

Figure 15. Cyclic Voltammogram of complex 6, [(2)AgCl].

Figure 16. Differential Pulse Voltammogram of Complex 6, [(2)AgCl].
Figure 17. Cyclic Voltammogram of complex 7, [(3)AgCl].

Figure 18. Differential Pulse Voltammogram of Compound 7, [(3)AgCl].
Figure 19. Cyclic Voltammogram of complex 10, [(2)₂Ag][Cl].

Figure 20. Differential Pulse Voltammogram of Complex 10, [(2)₂Ag][Cl].
Figure 21. Cyclic Voltammogram of complex 11, [(3)Ag][Cl].

Figure 22. Differential Pulse Voltammogram of Complex 11, [(3)Ag][Cl].
Figure 23. Cyclic Voltammogram of complex 12, [(4)2Ag][Cl].

Figure 24. Differential Pulse Voltammogram of Complex 12, [(4)2Ag][Cl].
\(^1\)H NMR and \(^{13}\)C NMR Spectra

**Figure 25.** \(^1\)H NMR spectrum (300 MHz, CDCl\(_3\)) of compound 7, [(3)AgCl].

**Figure 26.** \(^{13}\)C NMR spectrum (75 MHz, CDCl\(_3\)) of compound 7, [(3)AgCl].
Figure 27. $^1$H NMR spectrum (300 MHz, CDCl$_3$) of compound 9, [(1)Ag][Cl].

Figure 28. $^{13}$C NMR spectrum (75 MHz, CDCl$_3$) of compound 9, [(1)Ag][Cl].
Figure 29. $^1$H NMR spectrum (300 MHz, CDCl$_3$) of compound 10, [(2)$_2$Ag][Cl].

Figure 30. $^{13}$C NMR spectrum (75 MHz, CDCl$_3$) of compound 10, [(2)$_2$Ag][Cl].
Figure 31. $^1$H NMR spectrum (300 MHz, CDCl$_3$) of compound 11, [(3)Ag][Cl].

Figure 32. $^{13}$C NMR spectrum (75 MHz, CDCl$_3$) of compound 11, [(3)Ag][Cl].
Figure 33. $^1$H NMR spectrum (300 MHz, MeOD) of compound 12, [(4)$_2$Ag][Cl].

Figure 34. $^{13}$C NMR spectrum (75 MHz, MeOD) of compound 12, [(4)$_2$Ag][Cl].
**UV-Vis Absorption Spectra**

Figure 35. Electronic absorption spectra of complex 6 and 7 recorded in DMSO.

Figure 36. Electronic absorption spectra of complexes 10 and 11 recorded in DMSO.
Figure 37. Electronic absorption spectra of complex 12, [(4)\text{Ag}][\text{Cl}], 12 recorded in DMSO.
Infrared Spectroscopy Spectra

Figure 38. IR spectrum of complex 12, [(4)2Ag][Cl] recorded in DMSO.
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