Journal of the Korean Chemical Society
2021, *Vol. 65, No. 3*
Printed in the Republic of Korea
https://doi.org/10.5012/jkcs.2021.65.3.203

Synthesis, Characterization and *in vitro* Antibacterial Studies on Mixed Ligand Complexes of Iron(III) Based on 1,10-phenanthroline

Getinet Tamiru Tigineh*, Getu Sitotaw, Amogne Workie†, and Atakilt Abebe
Bahir Dar University, Science College, Chemistry Department, P.O. Box; 79, Bahir Dar, Ethiopia.
**E-mail: getenet.tamiru@bdu.edu.et; getsh2007@gmail.com*
†Institute of Chemistry, Academia Sinica, 128, Sec. Academia Rd., Nangang, Taipei 115, Taiwan.
(Received February 7, 2021; Accepted March 3, 2021)

ABSTRACT. As part of our attempt to discover novel active compounds against multi-drug resistant pathogens, we hereby report two new complexes of iron(III) with formulae: $[Fe(\mathbf{L}_1)_2(H_2O)_2]Cl_3$ and $[Fe(\mathbf{L}_1)_2(\mathbf{L}_2)(H_2O)]Cl_2$ where $\mathbf{L}_1$ = 1,10-phenanthroline ($C_{12}H_8N_2$) and $\mathbf{L}_2$ = guanide ($C_5H_4N_5O^-$). The synthesized complexes were characterized using spectroscopic analysis (ESI-MS, ICP-OES, FT-IR, and UV-Vis), cyclic voltammetry, CHN analysis, gravimetric chloride determination, melting point determination, and conductance measurement. Octahedral geometries are assigned to both complexes. *In vitro* antibacterial activity was tested on two Gram-positive (*Staphylococcus aureus, Streptococcus epidermidis*) and two Gram-negative (*Escherichia coli and Klebsiella pneumoniae*) bacteria using the disc diffusion method. The complexes demonstrated appreciable activity against these pathogens. Interestingly, the $[Fe(\mathbf{L}_1)_2(\mathbf{L}_2)(H_2O)]Cl_2$ complex manifested a higher degree of inhibition against the drug-resistant Gram-negative bacteria than the commercially available drug, namely erythromycin.

Key words: Iron(III), Mixed ligand complexes, 1,10-Phenanthroline, Guanide, Antibacterial activity

INTRODUCTION

Drugs derived from transition metals have been used for treating many infectious diseases[1] including cancer,[2] infections,[3] and inflammation.[4] The dynamic properties of transition metal compounds such as stability, redox, or coordination have been of interest, especially for multi-drug-resistant bacteria,[5–8] where natural product-based drugs become ineffective.[9–10] Quick penetrating in the cell membrane[11–12] or strong coordination to the DNA strand enhances the efficiency of metal-based drugs.[13–15]

The use of different ligands having different structures and properties is the primary strategy in tuning the properties of transition metal ions to obtain the desired applications.[16–18] Among various transition metals, Fe(III) is stable under physiological conditions and results in a thermodynamically stable complex. In particular, its mixed-ligand complexes have attracted growing attention for their biological activities. Especially, its complexes with aromatic heterocyclic ligands showed promising antibacterial activity.[12,19]

Nevertheless, any report on the synthesis and biological application in the form of mixed-ligand complexes containing flat heterocyclic ligands with nucleobases could not be found.

A large number of biologically active pharmaceutical ingredients containing flat heterocyclic ligands are commonly used in a wide variety of therapeutic areas. 1,10-Phenanthroline, among flat heterocyclic ligands, is a superb chelating bidentate ligand due to its ideally placed nitrogen atoms in the π-acidic rigid electron-poor heteroaromatic planar structure (*Fig.* 1). These properties endowed it with stacking interaction ability with DNA base pairs. Furthermore, when coordinated to metal ions, its complexes experience ionic or covalent or both interactions through the metal center with the organic base residues of the genetic materials.[20–22] Guanine is an oxypurine heteroaromatic organic base. It is a component of DNA and capable of interacting with cytosine residue of the genetic material of the pathogen, which interrupts the normal growth of bacteria (*Fig.* 1).[23]

Previously, our group reported ruthenium complexes with similar ligands for biological applications.[24] However, ruthenium is rare and expensive, whereas iron is compara-

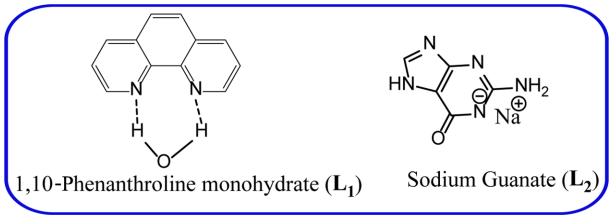


***Figure* 1.** Ligands used in this work.

tively abundant and cheap.[25] In terms of health benefits also iron is physiologically more relevant than ruthenium. These properties make us interested in the development of iron-based drugs along with ruthenium for multi-drug resistant pathogens.

Therefore, in this study, the structural investigation and biological activities of new iron complexes in a 1:2 Fe(III) to phen ($\mathbf{L}_1$) ratio alone as well as mixed with guanide ($\mathbf{L}_2$) in a 1:2:1 Fe(III) to phen ($\mathbf{L}_1$) to guanide ($\mathbf{L}_2$) are examined. The composition would result in a rigid three-dimensional structure and orchestrate the binding ability of iron(III) with a range of pathogen molecules and hinders their proliferation in the body of the host organism. As a result, the complexes would be a potential alternative drug for drug-resistant pathogens.

EXPERIMENTAL

Synthesis

New mononuclear Iron(III) complexes containing 1,10-phenanthroline alone and mixed with guanide were synthesized under optimized reaction conditions (*Scheme* 1).

Diaquabis(1,10-phenanthroline)ferrate(III) chloride $[Fe(L_1)_2(H_2O)_2]Cl_3$(1)

35 mL of methanolic solution of 1,10-phenanthroline monohydrated (0.3605 g, 2 mmol) was very slowly added from a buret to a 35 mL methanolic solution of $FeCl_3$ (0.1623 g, 1 mmol) magnetically stirred in 100 mL round bottom flask at room temperature. The reaction progress was monitored by TLC and the mixture was stirred for 2 h. Finally, a brown homogeneous solution was obtained. The solvent was removed at reduced pressure using a rotary evaporator. A brown powder was collected and washed and recrystallized from methanol.

mp; 160–162 °C; UV-Vis (Methanol, nm): 221 π→π* (C=C), 268 n→π*(C=N), 480, 512, 670 (d-d transition bands); FT-IR (KBr pellet, cm^{-1}): 3395 ν_{O-H}, 1543 $\nu_{C=N}$; ESI MS (Methanol, m/z): calculated for $[Fe(L_1)_2\text{-}H^+]$:415.27 $[M\text{-}H]^+$; found: 415.45; Anal. Calc. for $FeC_{24}H_{20}N_4O_2Cl_3$: C, 51.60; H, 3.61; N, 10.03; Cl, 19.06; Fe, 9.82. Found: C, 51.55; H, 3.58; N, 9.66; Cl, 19.03; Fe, 9.99. Molar conductivity: 234 (in H_2O) and 78 (in nitrobenzene) Scm^2mol^{-1}; Color: Brown; Yield: (0.4778 g, 86%).

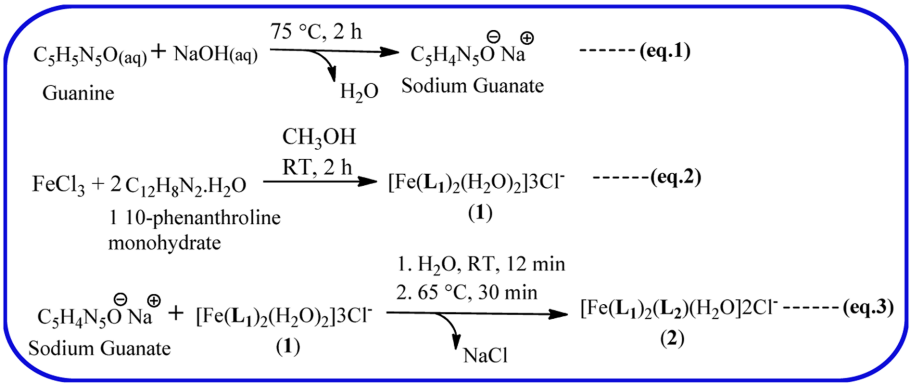
Aquaguanidebis(1,10-phenantroline)ferrate(III) chloride $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ (2)

A solution of guanide (0.150 g, 1 mmol), that was formerly prepared by reacting a hot aqueous solution of a mmol of guanine with NaOH (0.041 g, 1 mmol) at 75 °C, was added from a dropping funnel to an aqueous solution of complex 1 (0.559 g, 1 mmol) in a 100 mL round-bottomed flask being magnetically stirred. The mixture was stirred for 12 min and a break red homogeneous solution was obtained. The solution was refluxed at 65 °C for 30 min. Finally, a pink precipitate was formed, and the mixture was left to stand overnight at room temperature. The precipitate was separated from the solution phase using filter-paper and washed three times with double distilled water.

mp; >300 °C; UV-Vis (Methanol, nm): 270 n→π* (C=N), 420, 516, 739 (d-d transition bands); FT-IR (KBr pellet, cm^{-1}): 3460 ν_{O-H}, 1451 $\nu_{C=N}$; ESI MS (Methanol, m/z): calcd for $[Fe(L_1)_2+Na^+]$:439.30 $[M+Na]^+$; found: 439.45; Anal. Calc. for $FeC_{29}H_{22}N_9O_2Cl_2$: C, 53.15; H, 3.38; N, 19.24; Cl, 10.84; Fe, 8.53. Found: C, 53.01; H, 3.28; N, 18.98; Cl, 10.55; Fe, 8.22. Molar conductivity: 91.4 (in H_2O) and 57 (in nitrobenzene) Scm^2mol^{-1}; Color: Pink; Yield: (0.1880 g, 77%).

Antibacterial Activity Testing

In vitro antibacterial activities of the compounds against strains of two Gram-positive (*S. aureus* (ATCC25923) and *S.*



***Scheme* 1.** Synthesis strategy of Fe(III)-complexes.

epidermidis (ATCC12228) and two Gram-negative *E. coli* (ATCC25922) and *K. pneumoniae* (ATCC986605) bacteria were investigated by disc diffusion methods. Muller Hinton agar (MHA) and nutrient blood agar (BA) were used for culturing the bacterial isolates while diagnostic sensitivity test agar (oxoid Ltd BASINGSTOKE England) was used for sensitivity. All plates and Nutrient Broth (Difco) were autoclaved for 30 min at 121 °C in a steam sterilizer. The compounds dissolved in double-distilled water were applied at a fixed concentration of 500 μg/mL. Zones of inhibition were measured after 24 h incubation at 37 °C. Antibiotic discs Erythromycin was used as a reference. The minimum inhibitory concentration (MIC) of the two complexes against each bacterium was determined by preparing different concentrations of the complexes by serial dilution (75 μg/mL, 150 μg/mL, 300 μg/mL, 500 μg/mL, and 600 μg/mL). The antibacterial tests were performed at the Amhara Regional Health Research Microbiology Laboratory Center, Bahir Dar, Ethiopia.

RESULTS AND DISCUSSION

The analytical data of the complexes agree well with the assigned molecular formulae of complexes **1** and **2**. The gravimetric chloride determination confirmed the presence of three and two chloride ions in the outer sphere of complexes **1** and **2**, respectively. The molar conductivity values of complexes **1** and **2** revealed their electrolytic nature. Their conductivities were found higher in an aqueous solution than in nitrobenzene. The result is in close agreement with the literature.[26] ICP-OES metal determination showed the anticipated amount of iron in complexes. Furthermore, the experimental and theoretical data of C, H, and N elemental analyses for complexes are in good agreement, which confirms the achievement of the intended complexes (*see experimental section*).

FT-IR Spectroscopy

The infrared spectra of the ligands and the complex are indicated in *Fig.* S1 and selected characteristic frequencies are listed in *Table* S1. The sharp band at 3430 cm^{-1} (s), characteristic for ν(OH) stretching in the free 1,10-phenanthroline monohydrate appeared broad at 3395 cm^{-1} (m) in $[Fe(L_1)_2(H_2O)_2]Cl_3$. This change in the absorption frequency of water explains the change like its interaction, consequently confirmed the transformation of crystalline H_2O to coordinated H_2O. The band arising due to vibrational ν(C=N) mode at 1588 cm^{-1}(m) in the free 1,10-phenanthroline was shifted to lower frequencies, at 1543 cm^{-1}(m) in $[Fe(L_1)_2(H_2O)_2]Cl_3$. This shift suggested that 1,10-phenanthroline binds with the metal center *via* ring nitrogen.

The presence of the characteristic peaks, in the region of 1490–1475 cm^{-1}, of guanine in $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ attributed to the successful coordination of guanide to the metal center. Besides, the coordination of guanide with metal *via* the imidazole ring nitrogen was confirmed by the shift in ν(C=N) vibration.[27–29] Furthermore, the similarity in the stretching frequency of ν(C=O) and scissoring of δ(NH_2) in free guanine with that of the stretching frequency of ν(C=O) and scissoring of δ(NH_2) in $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ respectively confirmed oxygen of the carbonyl group (C=O) and nitrogen of amine (NH_2) group of guanide are not used for coordination.[27]

The two sharp peaks at frequencies 3118 and 3320 cm^{-1} are assigned to symmetrical and antisymmetric ν(NH_2) stretching-modes.[30] These were shifted to 3108 and 3344 cm^{-1} (b) in $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ *Fig.* 2. It is worth mentioning that because of its special structure and the position of the double bonds in the rings, guanide is presumably subjected to electron delocalization. This may result in shifts in ν(NH_2) vibration due to an increase in the sp^2 character of the amino nitrogen atom[31] upon coordination with imidazole ring nitrogen.

The changes in absorption frequencies and peak strength together with the appearance of non-ligand bands on the complexes suggested that 1,10-phenanthroline and guanide are successfully coordinated to the metal center.

Electronic Absorption Spectroscopy

The electronic spectra of the complexes run in aqueous solutions are displayed in *Table* S2 and *Fig.* S2. The aqueous solution of $FeCl_3$ results in slightly elongated octahedral $[Fe(H_2O)_6]Cl_3$. The band at 300 nm represents LMCT from

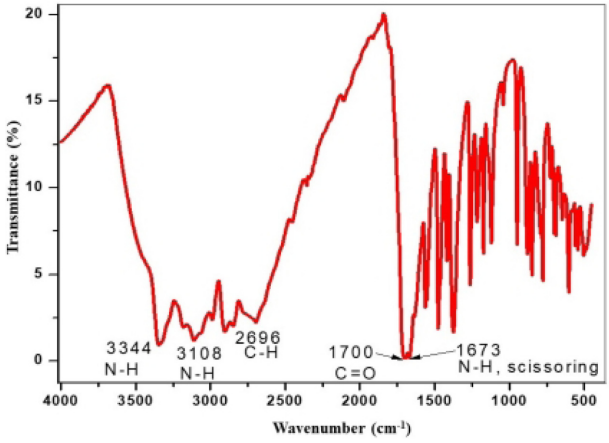


***Figure* 2.** FT-IR spectra of $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ (**2**).

the electron-rich H_2O orbitals to the low-lying all singly occupied Fe(III) t_{2g} orbitals. In complex **1**, the coordination of the electron-deficient, bidentate strong field 1,10-phenanthroline in the equatorial position results in a short but strong bond. This results in the lowering of the d_{z^2} as well as the population of all the d electrons in the t_{2g} orbitals of Fe(III). Consequently, the LMCT takes place in the low-lying d_{z^2} orbitals. This fact is signaled by the band at 353 nm. The increase in the LMCT resonance wavelength indicates the lower energy gap between the donor ligand and recipient metal orbitals.[32] The non-ligand bands newly observed in complexes **1** and **2** in the range of 13333–23809 cm^{-1} corresponding to the transitions $^2T_{2g}\rightarrow{}^2A_{2g}$, $^2T_{2g}\rightarrow{}^2T_{1g}$, $^2T_{2g}\rightarrow{}^2E_g$ confirm the possible coordination of metal and ligands.[33]

MS Spectroscopy

The ESI MS spectra of the complexes recorded dissolving in methanol showed a characteristic molecular ion peak of $[Fe(L_1)_2(H_2O)_2]Cl_3$ with the best accuracy at m/z calculated for $[Fe(L_1)_2\text{-}H^+]$:415.27 $[M\text{-}H]^+$; found: 415.45. This is presumably due to the electron-deficient $[Fe(L_1)_2]^{3+}$ complex captured low-energy electron generated in the ionization chamber and undergone reduction to $[Fe(L_1)_2]^{2+}$ before deprotonation.[34] Alternatively, the observed unique fragmentation patterns of $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ at m/z = 439.3 and 465.3, which represent the molecular ion peaks of $[Fe(L_1)_2{+}Na^+]$[35] and $[Fe(L_1)_2(H_2O)(CH_3OH)\text{-}H^+]$,[36] respectively, and the multiple-deprotonated species at 563.2, and 383.1 corresponds to $[Fe(L_1)_2(L_2)\text{-}3H^+]$ and $[Fe(L_1)(L_2)\text{-}3H^+]$ respectively[37] *Fig.* S3, windup the shreds of evidence found in the former techniques in confirming the achievement of the intended complexes.

Cyclic Voltammetry

Fig. S4 shows the CVs of $[Fe(L_1)_2(H_2O)_2]Cl_3$ and $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ for successive cycles at a scan rate of 100 mVs^{-1}. The voltammogram displays a cathodic peak at the potentials 0.84 and 0.84 V and an anodic peak at 0.87 and 0.86 V of $[Fe(L_1)_2(H_2O)_2]Cl_3$ and $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$, respectively. The pair of peaks in both complexes corresponds to the reversible redox process of the species as Fe(II) and Fe(III) system.[38] The absence of peaks other than expected cathodic and anodic peaks for the Fe(II) and Fe(III) system signifies that the complexes are stable in the voltage range scanned here. These results conform to the literature values.[39]

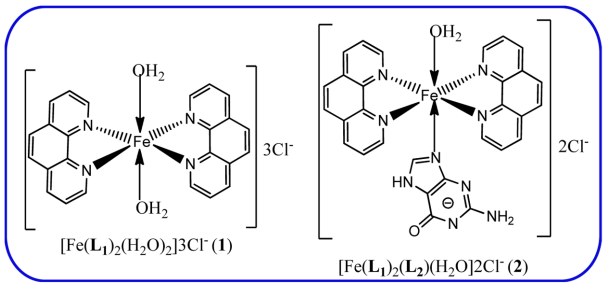


***Figure* 3.** Proposed structures of complexes.

Antimicrobial Activity

***Antibacterial Activity Testing*:** The examination showed that the complexes demonstrated biological activities against all tested strains (*Fig.* 4 and *Table* S3).

The observed increase in antibacterial activity compared to the ligands can be based on Overton's concept[40] and Tweedy's chelation theory.[41] An important condition for the antimicrobial activity of a compound is its ability to pass through the lipid membrane that surrounds the cell. On coordination, the polarity of the metal ion will be reduced to a greater extent due to the overlap of the ligand orbitals and partial sharing of the positive charge of the metal ion with the donor groups, which significantly increases the lipophilicity of the complex. This increased liposolubility enhances the penetration of the complexes into the lipid membrane and interferes with the normal activities of the bacteria.[40-43]

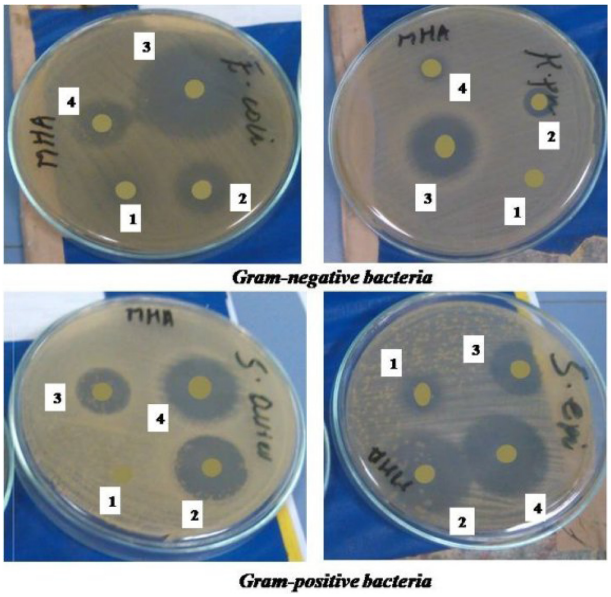


***Figure* 4.** Inhibition zone at 500 μg/mL concentration of (1) metal salt ($FeCl_3$), (2)$[Fe(L_2)_2(H_2O)_2]Cl_3$, (3) $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ and (4) commercial antibiotic (erythromycin) against Gram-positive (Staphylococcus aureus, Streptococcus epidermidis) and Gram-negative (K. pneumoniae and E. coli) bacteria.

The two newly synthesized Fe(III)-complexes manifested two interesting phenomena compared with commercially available drug erythromycin. A comparative study verified that $[Fe(L_1)(H_2O)_2]Cl_3$ showed virtually equal activity to erythromycin against Gram-positive bacteria, whereas $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ demonstrated a notably high antibacterial activity than erythromycin even against Gram-negative bacteria. The latter observation is crucial as Gram-negative bacteria are highly drug-resistant due to their thick impenetrable cell wall and deadliest pathogens.[44] The better activities demonstrated by $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ against Gram-negative bacteria compared to $[Fe(L_1)_2(H_2O)_2]Cl_3$ are presumably due to its additional interaction with the cytosine residue of the genetic material of the cell by guanide.[23] The observed biological activities of the complexes are different from those of the starting materials and the possible fragments, which suggested a unique characteristic of complexes and their inertness in the media. This argument is based on the strong field nature of the ligands coordinated to the metal.

Literature data revealed that Fe(II) complexes with acyclic chelating ligands demonstrated low or virtually no antibacterial activity against the tested Gram-negative and Gram-positive bacterial.[45-46] This shows a mixed ligand Fe(III) complexes of aromatic heterocyclic chelating ligands and nucleobase are promising combinations to develop new metal-based drugs with better antibacterial activity. The small ionic radii of the Fe(III) ion comparing to Fe(II) together with the aforementioned mixed ligands effect explains the exceptional antibacterial activity due to the ease of penetration of Fe(III) complexes of the cell membrane.[12] A comparative study of the latter complexes with clinical drugs such as Gentamycin[17] and Ciprofloxacin[18] also showed an analogous or even higher antibacterial activity against the tested Gram-negative and Gram-positive bacterial depending on the ligand and metal. In our current investigation, erythromycin, which was used as a clinical drug for comparison, is an antibiotic obtained from the bacterial Streptomyces erythreus and is effective against many Gram-positive and some Gram-negative bacteria.[47]

The high antibacterial activity of $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ against Gram-negative bacteria compared to erythromycin, makes the complex potential alternative drug for treating diseases caused by Gram-negative bacteria after passing cytotoxicity testing.

***Minimum Inhibitory Concentration (MIC) Determination*:** The minimum inhibitory concentrations of $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ complex are listed in *Table* S4. Both complexes at 75 μg/mL were inactive against the whole bacterial species. As far as this specific condition is concerned, the lowest concentration of $[Fe(L_1)_2(L_2)(H_2O)]Cl_2$ complex that completely inhibited the growth of microorganisms after 24 h of exposure is 150 μg/mL for Gram-negative bacteria and 500 μg/mL for Gram-positive bacteria. The aforementioned complex has higher antibacterial activity for Gram-negative bacteria at lower concentrations than for Gram-positive bacteria.

CONCLUSION

In this work, we successfully synthesized new iron(III)-1,10-phenanthroline complex in 1:2 metal to ligand ratio and its mixed ligand complex with guanide. The physicochemical and spectroscopic data suggest that Fe(III) and the ligands are brought together with rigid configuration and hence both the complexes proposed having octahedral geometry (*Fig.* 3). The cyclic voltammetry study confirmed the redox stability of the synthesized complexes. The antibacterial test showed a high degree of inhibition by the complex with guanide against drug resisting Gram-negative bacteria. The results provide new insights on metal-based drugs, particularly coordinated with nucleobases, thus the overall biological findings of this work would be useful in improving the effectiveness of therapeutic drugs. Based on the observations, the *in vivo* shall be investigated, as a continuation of this study.

Competing Interests. There is no conflict of interest between the authors and the funding institution.

Acknowledgments. The authors express sincere gratitude to Bahir Dar University for providing the necessary facilities. We thank Dr.Yonas Beyene for running CV at the electroanalytical laboratory, Bahir Dar University, Ethiopia. We also thank Academia Sinica, Institute of Chemistry, for allowing us to run ESI-MS and EA in Taiwan. And the Publication cost of this paper was supported by the Korean Chemical Society.

Supporting Information. Additional supporting information may be found online in the Supporting Information section at the end of the article.

REFERENCES

1. Rafique, S.; Idrees, M.; Nasim, A.; Akbar, H.; Athar, A. *Biotechnol. Mol. Biol. Rev.* **2010**, *5*, 38.
2. Abu-Dief, A. M.; Abdel-Rahman, L. H.; Shehata, M. R.;

Abdel-Mawgoud, A. A. H. *J. Phys. Org. Chem.* **2019**, *32*, e4009.
3. Mukherjee, T.; Pessoa, J. C.; Kumar, A.; Sarkar, A. R.; *Dalton Trans.* **2013**, *42*, 2594.
4. Weder, J. E.; Dillon, C. T.; Hambley, T. W.; Kennedy, B. J.; Lay, P. A.; Biffin, J. R.; Davies, N. M. *Coord. Chem. Rev.* **2002**, *232*, 95.
5. Hambley, T. W. *Dalton Trans.* **2007**, *43*, 4929.
6. Ronconi, L.; Sadler, P. J. *Coord. Chem. Rev.* **2007**, *251*, 1633.
7. Graf, N.; Lippard, S. J. *Adv. Drug Deliv. Rev.* **2012**, *64*, 993.
8. Ming, L. J. *Med. Res. Rev.* **2003**, *23*, 697.
9. Al Qaraghuli, M. M.; Alzahrani, A. R.; Niwasabutra, K.; Obeid, M. A.; Ferro, V. A. *Ann. Pharm. & Pharm. Sci.* **2017**, *2*, 1.
10. David, B.; Wolfender, J. L.; Dias, D. A. *Phytochem Rev.* **2015**, *14*, 299.
11. Turel, I. *Coord. Chem. Rev.* **2002**, *232*, 27.
12. Hassoon, A. A.; Harrison, R. G.; Nawar, N.; Smith, S. J.; Mostafa, M. M. *J. Mol. Struct.* **2020**, *1203*, 127240.
13. Fleisher, M. B.; Waterman, K. C.; Turro, N. J.; Barton, J. K. *Inorg. Chem.* **1986**, *25*, 3549.
14. Long, E. C.; Barton, J. K. *Acc. Chem. Res.* **1990**, *23*, 271.
15. Prabhakara, M. C.; Basavaraju, B.; Naik, H. S. *Bioinorg Chem. Appl.* **2007**, *2007*.
16. Bryan Sears, R.; Joyce, L. E.; Turro, C. *Photochem. Photobiol.* **2010**, *86*, 1230.
17. Tamiru, G.; Abebe, A.; Abebe, M.; Liyew, M. *Ethiop. J. Sci. & Technol.* **2019**, *12*, 69.
18. Abebe, A.; Bayeh, Y.; Belay, M.; Gebretsadik, T.; Thomas, M.; Linert, W. *Future J. Pharm. Sci.* **2020**, *6*, 1.
19. Soliman, S. M.; Al-Rasheed, H. H.; Albert, J. H.; El-Faham, A. *Molecules* **2020**, *25*, 5750.
20. Reed, J. E.; White, A. J.; Neidle, S.; Vilar, R. *Dalton Trans.* **2009**, *14*, 2558.
21. Melnic, E.; Coropceanu, E. B.; Kulikova, O. V.; Siminel, A. V.; Anderson, D.; Rivera-Jacquez, H. J.; Masunov, A. E.; Fonari, M. S.; Kravtsov, V. C. *J. Phys. Chem. C.* **2014**, *118*, 30087.
22. Abebe, A.; Atlabachew, M.; Liyew, M.; Ferede, E. *Cogent Chem.* **2018**, *4*, 1476077.
23. Shapiro, R. In *Progress in Nucleic Acid Research and Molecular Biology*; 1968; Academic Press: Vol. 8, pp 73-112.
24. Abebe, A.; Hailemariam, T. *Bioinorg. Chem. Appl*. **2016**, 3607924.
25. Wenger, O. S. *Chem. Eur. J.* **2019**, *25*, 6043.
26. Ali, I.; Wani, W. A.; Saleem, K. *Synthesis and Reactivity in Inorganic, Metal-Organic, and Nano-Metal Chemistry* **2013**, *43*, 1162.
27. Mikulski, C. M.; Mattucci, L.; Smith, Y.; Tran, T. B.; Karayannis, N. M. *Inorg. Chim. Acta* **1983**, *80*, 127.
28. Mastropietro, T. F.; Armentano, D.; Grisolia, E.; Zanchini, C.; Lloret, F.; Julve, M.; De Munno, G. *Dalton Trans.* **2008**, *4*, 514.
29. Sheina, G. G.; Stepanian, S. G.; Radchenko, E. D.; Blagoi, Y. P. *J. Mol. Struct.* **1987**, *158*, 275.
30. Mathlouthi, M.; Seuvre, A. M.; Koenig, J. L. *Carbohydr. Res.* **1986**, *146*, 15.
31. Chandra, A. K.; Nguyen, M. T.; Uchimaru, T.; Zeegers-Huyskens, T. *J. Phys. Chem. A* **1999**, *103*, 8853.
32. Lever, A. P. Inorganic Electronic Spectroscopy 2nd ed., Elsevier: Amsterdam, 1984.
33. Lobanov, S. S.; Hsu, H.; Lin, J. F.; Yoshino, T.; Goncharov, A. F. *J. Geophys. Res., Solid Earth* **2017**, *122*, 3565.
34. Nicolescu, T. O. *Mass Spectrometry* **2017**, 24.
35. Looi, D. W.; Eyler, J. R.; Brajter-Toth, A. *Electrochim. Acta* **2011**, *56*, 2633.
36. Cheng, P.; Li, Y.; Li, S.; Zhang, M.; Zhou, Z. *Phys. Chem. Chem. Phys.* **2010**, *12*, 4667.
37. Banerjee, S.; Mazumdar, S. *Int. J. Anal. Chem.* **2012**, *2012*.
38. Zhang, Q. L.; Liu, J. G.; Chao, H.; Xue, G. Q.; Ji, L. N. *J. Inorg. Biochem.* **2001**, *83*, 49.
39. de Castro, M. L.; Valcarcel, M.; Albahadily, F. N.; Mottola, H. A. *J. Electroanal. Chem.* **1987**, *219*, 139.
40. Anjaneyulu, Y.; Rao, R. P. *Synth. React. Inorg. Met. Org. Chem.* **1986**, *16*, 257.
41. Tweedy, B. G. *Phytopathology* **1964**, *55*, 910.
42. Warra, A. A. *J. Chem. Pharm. Res.* **2011**, *3*, 951.
43. Mohamad, A. D. M.; Abualreish, M. J. A.; Abu-Dief, A. M. *J. Mol. Liq.* **2019**, *290*, 111162.
44. Moon, M. S. *Asian Spine J.* **2019**, *13*, 343.
45. Mahmud, T.; Rehman, R.; Gulzar, A.; Khalid, A.; Anwar, J.; Shafique, U.; Salman, M. *Arab. J. Chem.* **2010**, *3*, 219.
46. Lawal, A.; Shodeinde, A. S.; Amolegbe, S. A.; Elaigwu, S. E.; Yunus-Issa, M. T. *J. Appl. Sci. Environ. Manage.* **2017**, *21*, 568.
47. World Health Organization. *WHO Model Prescribing Information: Drugs Used in Skin Diseases*; World Health Organization; Geneva, 1997; 1-126.