Science Journal of Chemistry
2020; 8(1): 1-6
http://www.sciencepublishinggroup.com/j/sjc
doi: 10.11648/j.sjc.20200801.11
ISSN: 2330-0981 (Print); ISSN: 2330-099X (Online)

Synthesis, Characterization and Bacterial Growth Inhibitory Properties of Schiff-Base Ligands Derived from Amino Acids

James Tembei Titah[1, *], Coulibaly Wacothon Karime[2], Kevin Chambers[1], Anita Balogh[1], Kevin Joannou[1]

[1]Department of Mathematical and Physical Sciences, Concordia University of Edmonton, Chemistry Research Laboratory, Edmonton, Canada

[2]Department of Chemistry and Biochemistry, Peleforo Gon Coulibaly University, Korhogo, Ivory Coast

Email address:

tt.james@unb.ca (J. T. Titah), james.titah@rdc.ab.ca (J. T. Titah), james.titah@concordia.ab.ca (J. T. Titah), tijames2001@yahoo.com (J. T. Titah)

[*]Corresponding author

To cite this article:

James Tembei Titah, Coulibaly Wacothon Karime, Kevin Chambers, Anita Balogh, Kevin Joannou. Synthesis, Characterization and Bacterial Growth Inhibitory Properties of Schiff-Base Ligands Derived from Amino Acids. *Science Journal of Chemistry.* Vol. 8, No. 1, 2020, pp. 1-6. doi: 10.11648/j.sjc.20200801.11

Received: January 23, 2020; **Accepted**: February 13, 2020; **Published**: March 2, 2020

Abstract: Schiff-base ligands and their metal complexes are attracting a lot of research in bioinorganic and medicinal chemistry owing to their improved activity in biological systems. Six schiff-base ligands derived from amino acids; *N*-Salicylidene Alanine, *N*-Salicylidene Serine, *N*-Benzalidene Histidine, *N*-Balzalidene Leucine, *N*-4-(dimethylamino) benzalidene Phenylalanine, and *N*-4-(dimethylamino)benzalidene Valine have been synthesized, characterized and their bacterial growth inhibitory properties determined against *Staphylococcus aureus* and *Escherichia coli*. These schiff-bases are synthesized by the condensation reaction between carbonyl compounds (aldehydes and ketones) and amines (amino acids). Characterization of the schiff-base ligands is done using melting/decomposition temperatures, FTIR spectroscopy, US-visible spectroscopy, and solubility. It is observed that, all the schiff-base ligands contain the imine or azomethine (C=N) group with a stretching frequency ranging from 2200 - 2400 cm^{-1}. In addition, all the schiff-base ligands are seen to be soluble in water, which is paramount in their application in biological systems. The structures of the schiff-base ligands were deduced based on the characterization techniques. Furthermore, the bacterial growth inhibitory properties of the schiff-base ligands were done using the Agar Well Diffusion method. The results reveal that, all the schiff-base ligands show no toxicity effect or negative bacterial growth properties against gram positive (*Staphylococcus aureus*) and gram negative (*Escherichia coli*) bacteria.

Keywords: Amino Acids, Characterization, Schiff-Bases, Synthesis, Biological Activities, Imine Group

1. Introduction

Schiff-base ligands and their bio-active metal complexes have been studied extensively for the past decades for applications in bioinorganic and medicinal chemistry, and the research is ongoing. Schiff-bases provide potential binding sites for bio-chemically active molecules and are generally synthesized by the condensation reaction between amines and carbonyl compounds (aldehydes and ketones). In addition, schiff-bases are recognized by the presence of an imine or azomethine (C=N) group [1-9]. The presence of nitrogen, oxygen and/or sulphur donor atoms in schiff-bases play an important role in biological systems and can move across the phospholipid layers of membrane by active transport. Schiff-base ligands and their metal complexes have been used industrially as catalysts in the presence of moisture and exhibit a wide range of applications including biological activities such as; anti-fungal, anti-malarial, anti-bacterial, anti-diabetic, anti-cancer, anti-proliferative, anti-inflammatory, anti-viral, anti-tumor, etc properties. Development of new chemotherapeutic schiff-base ligands and their metal complexes is now attracting a lot of attention

in Bioinorganic and Medicinal Chemistry [10-17]. This paper will present the syntheses and characterization of six schiff-base ligands derived from amino acids (alanine, serine, phenylalanine, histidine, leucine and valine) as a preliminary step towards synthesising their metal complexes.

Characterization will include techniques such FTIR, UV-Visible spectroscopic techniques, melting/decomposition temperatures, solubility tests, etc. Furthermore, the bacterial growth inhibitory properties of the ligands will be determined against Gram positive and Gram negative bacteria; *Staphylococcus aureus* and *Escherichia coli* respectively. Since these ligands and/or their metal complexes are intended for use in biological systems, the study of their toxicity effects on bacteria is paramount for the continuation in synthesizing and characterizing the metal complexes [14, 15]

2. Experimentation

2.1. General Synthetic Method

An equimolar amount of NaOH pellets and the amino acid are dissolved in a beaker using a suitable solvent (methanol/ethanol) with the help of a magnetic stirrer at room temperature. An equimolar amount of the carbonyl compound (salicylaldehyde and 4-dimethyaminobenzaldehyde) was dissolved in a methanol/ethanol and added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to go on for about 30-45 mins and then evaporated to about 50-70% of the original mixture. An equimolar amount of acetic acid was added to the mixture and allowed to stand for 1-3 hours or overnight. The resulting coloured crystals (powder) were filtered by suction, washed with methanol/ethanol mixture and air dried. The powder (crystals) was preserved in vials for subsequent analyses [16].

Agar Well Diffusion: Wells were bored into nutrient agar petri dishes. Each dish was streaked with a suspension of either *Staphylococcus aureus* or *Escherichia coli* (KL25) to produce a lawn of growth in the case of no inhibition. The wells were filled with a solution of one tested schiff-base ligand in DMSO per plate at a concentration of 10 mmols, with 5 wells per plate, therefore, 5 replicates of each treatment. For each bacteria species, one plate was negative a control (DMSO only) and one was positive a control (Streptomycin in DMSO). All plates were incubated at 37°C for 22 hours before measuring zones of inhibition.

Analysis: No statistical testing was done on results because all schiff-base treatments had identical averages and standard deviations to the negative control.

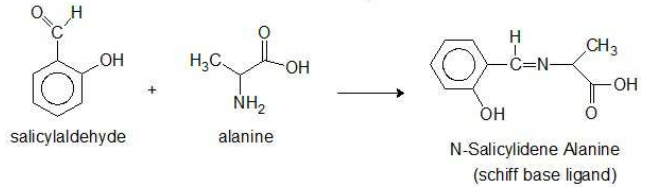

Figure 1. *General Synthetic method of the Schiff-Bases.*

2.1.1. Synthesis of N-Salicylidene Alanine (1)

Sodium hydroxide (0.075 mols) and alanine (0.075 mols) were dissolved in 30.0 mL methanol/ethanol mixture in a beaker and stirred continuously with the help of a magnetic stirrer at room temperature. An equimolar amount of salicylaldehyde (0.075 mols) dissolved in 30.0 mL methanol/ethanol mixture was added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to react for 45 mins and the solvent evaporated to 60% of the original mixture. Acetic acid (0.075 mols) was added to the mixture and allowed to stand overnight. The resulting yellow powder was filtered by suction, washed with 10 mL methanol/ethanol mixture and air dried. The powder was stored in a vial and the percentage yield was 85%. All the chemicals used in this research were used as purchased without any modification.

2.1.2. Synthesis of N-Salicylidene Serine (2)

Sodium hydroxide (0.060 mols) and serine (0.060 mols) were dissolved in 30.0 mL methanol/ethanol mixture in a beaker and stirred continuously with the help of a magnetic stirrer at room temperature. An equimolar amount of salicylaldehyde (0.060 mols) dissolved in 30.0 mL methanol/ethanol mixture was added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to react for one hour and the solvent evaporated to 50% of the original mixture. Acetic acid (0.060 mols) was added to the mixture and allowed to stand overnight. The resulting yellow powder was filtered by suction, washed with 10 mL methanol/ethanol mixture and air dried. The powder was stored in a vial and the percentage yield was 80%.

2.1.3. Synthesis of N-Benzalidene Histidine (3)

Sodium hydroxide (0.055 mols) and histidine (0.055 mols) were dissolved in 30.0 mL methanol/ethanol mixture in a beaker and stirred continuously with the help of a magnetic stirrer at room temperature. An equimolar amount of benzaldehyde (0.055 mols) dissolved in 40.0 mL methanol/ethanol mixture was added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to react for one hour and the solvent evaporated to 70% of the original mixture. Acetic acid (0.055 mols) was added to the mixture and allowed to stand overnight. The resulting yellow powder was filtered by suction, washed with 10 mL methanol/ethanol mixture and air dried. The powder was stored in a vial and the percentage yield was 80%.

2.1.4. Synthesis of N-Benzalidene Leucine (4)

Sodium hydroxide (0.050 mols) and leucine (0.050 mols) were dissolved in 30.0 mL methanol/ethanol mixture in a beaker and stirred continuously with the help of a magnetic stirrer at room temperature. An equimolar amount of benzaldehyde (0.050 mols) dissolved in 40.0 mL methanol/ethanol mixture was added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to react for one hour and the solvent evaporated to 70% of the original mixture. Acetic acid (0.050 mols) was added to the mixture and allowed to stand overnight. The resulting yellow powder was filtered by suction, washed with

10 mL methanol/ethanol mixture and air dried. The powder was stored in a vial and the percentage yield was 85%.

2.1.5. Synthesis of N-4-(dimethylamino)benzalidene Phenylalanine (5)

Sodium hydroxide (0.060 mols) and phenylalanine (0.060 mols) were dissolved in 30.0 mL methanol/ethanol mixture in a beaker and stirred continuously with the help of a magnetic stirrer at room temperature. An equimolar amount of 4-dimethylaminobenzaldehyde (0.060 mols) dissolved in 40.0 mL methanol/ethanol mixture was added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to react for one hour and the solvent evaporated to 60% of the original mixture. Acetic acid (0.060 mols) was added to the mixture and allowed to stand overnight. The resulting yellow powder was filtered by suction, washed with 10 mL methanol/ethanol mixture and air dried. The powder was stored in a vial and the percentage yield was 80%.

2.1.6. Synthesis of N-4-(dimethylamino)benzalidene Valine (6)

Sodium hydroxide (0.050 mols) and valine (0.050 mols) were dissolved in 30.0 mL methanol/ethanol mixture in a beaker and stirred continuously with the help of a magnetic stirrer at room temperature. An equimolar amount of 4-dimethylaminobenzaldehyde (0.060 mols) dissolved in 40.0 mL methanol/ethanol mixture was added drop wise to the resulting mixture while stirring continuously. The reaction was allowed to react for one hour and the solvent evaporated to 60% of the original mixture. Acetic acid (0.050 mols) was added to the mixture and allowed to stand overnight. The resulting yellow powder was filtered by suction, washed with 10 mL methanol/ethanol mixture and air dried. The powder was stored in a vial and the percentage yield was 75%.

2.2. Bacterial Growth Inhibitory Properties

The bacterial growth inhibitory properties of the schiff-base ligands were done using the Agar Well Diffusion method. The results reveal that, all the schiff-base ligands show no toxicity effect or negative bacterial growth properties against gram positive (*Staphylococcus aureus*) and gram negative (*Escherichia coli*) bacteria.

3. Results and Discussion

3.1. Melting Point/Decomposition Temperatures

The melting point/decomposition temperatures of all the schiff-base ligands and their corresponding amino acids were determined using the melting point apparatus. The schiff-bases were seen to melt at different temperatures compared to the amino acids from which they were synthesized, indicating formation of new products. The schiff-bases were observed to decompose at temperatures above 300°C. The results are presented in Table 1.

Table 1. *Melting point/decomposition temperatures (°C) of the Amino Acids and their corresponding Schiff Bases.*

Schiff-Bases	1	2	3	4	5	6
M. pt of amino acids (°C)	297	222	254	>300	273	298
M. pt of Schiff-Bases (°C)	193	167	267	268	207	198

3.2. FTIR Spectra of the Schiff-Bases

The FTIR spectra of all the schiff-bases were obtained using the FTIR instrument. It was observed that all the schiff-bases show the prominent imine (C=N) peak around 7324 cm^{-1}. The average IR stretching frequencies of all the schiff-bases are presented in Table 2 and the prominent peaks for compound 1 in Figure 2 [11].

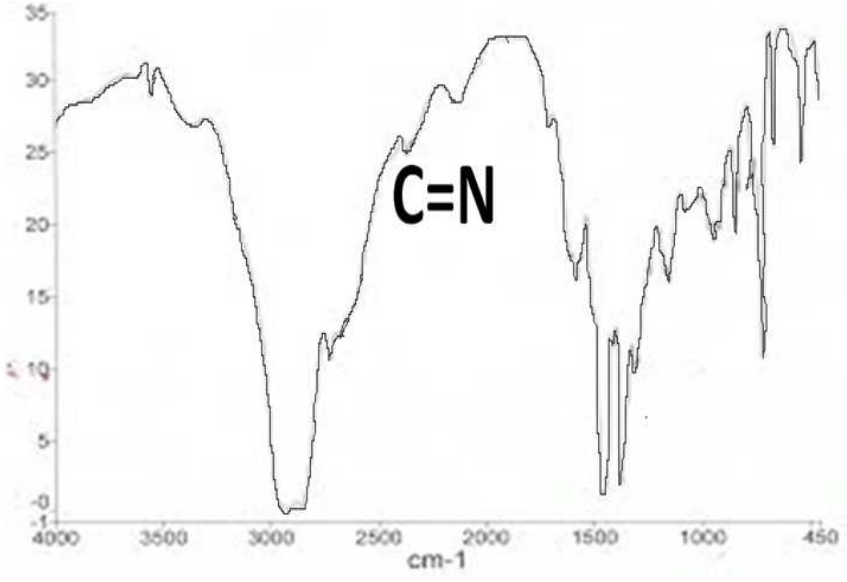

Figure 2. *IR spectra showing prominent C=N peak in compound 1.*

Table 2. *Average IR vibration frequencies showing the prominent peaks.*

Average IR stretching frequencies (cm^{-1})	IR stretching frequency ranges (cm^{-1})	Assignment
3555	>3500	Aromatic C-H stretching
3375	3400-3650	Alcohol O-H stretching
2905	2500-3100	Carboxylic acid O-H stretching
2374	2200-2400	Nitrile C=N (imine) stretching
1668	1650-1725	C=O stretching
1461	1450-1600	Aromatic C=C stretching
1377	1400-1600	Aromatic C=C bending
721	690-900	Aromatic C-H bending

3.3. UV-visible Spectroscopy and Energy Calculation

With the exception of *N*-Benzalidene Leucine (4), which is very pale yellow or almost colourless, all the schiff-bases were coloured compounds indicating that they will absorb light in the UV-visible region. The coloured schiff-bases showed a peak in the UV-visible region with maximum absorption wavelengths. The energies corresponding to the maximum absorption are similar indicating that the schiff-bases absorb in the same wavelength range in the UV-visible region. Table 3 shows the maximum energies of absorption of the schiff-bases in the UV-visible region.

Table 3. *Maximum Energies of Absorption in the UV-visible region. * very pale yellow.*

Schiff-base	1	2	3	4	5	6
Λmax (nm)	350	347	289	157	387	380
Energy (x 10-19J)	5.679	5.728	5.728	5.570	5.140	5.230
Colour	Yellow	Yellow	Pale-Yellow	Colourless*	Yellow	Yellow

3.4. Solubility

The solubility of the schiff-bases were performed in some solvents. It is of paramount importance to determine the solubility of the schiff-bases especially in water since they are intended for use in biological systems. This was done by dissolving 0.10 g sample of schiff-base in 5.00 mL of solvent and manually examining their solubilities. It is important to note that all the schiff-bases are soluble in water. The results of the solubility test are presented in Table 4.

Table 4. *Solubility of the schiff-bases in selected solvents.*

Solvent/Bases	1	2	3	4	5	6
Water (H_2O)	ss	s	s	S	s	S
Methanol	ss	ss	ss	Ss	ss	ss
Ethanol	ss	ss	ss	Ss	ss	ss
Chloroform	i	i	i	I	i	I
Hexane	i	i	i	I	i	I
D_2O	s	s	s	S	s	S

ss = sparingly soluble, s = soluble, i = insoluble.

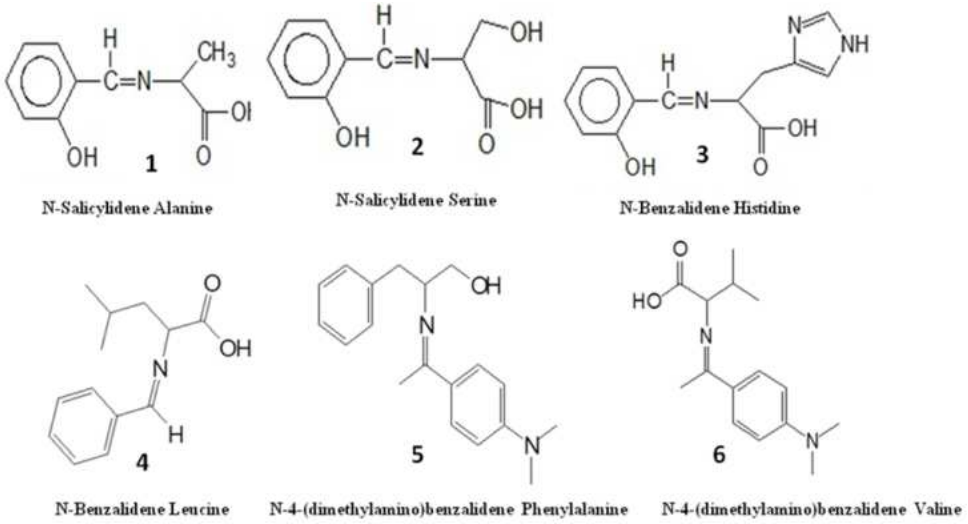

Figure 3. *Predicted Structures of the Schiff-Bases.*

4. Bacterial Growth Inhibitory Properties

The bacterial growth inhibitory properties of the six schiff-base ligands were tested against Gram positive and Gram negative bacteria; *Staphylococcus aureus* and *Escherichia coli* respectively. The results are presented in figures 4 and 5.

From the results presented in Figures 4 and 5, it is exceptionally clear that none of the schiff-bases showed any bacterial growth inhibitory properties or toxicity effects against *S. aureus and E. coli*. These preliminary results on the schiff-bases are very good and would be extended to their respective metal complexes in the next edition of this publication. We will also extend our studies to the testing of other biological activities to eukaryotes.

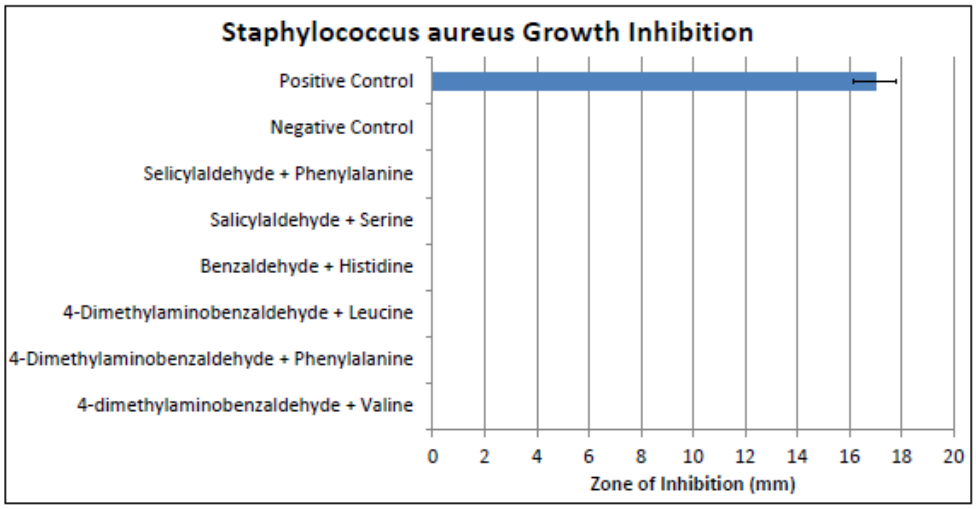

Figure 4. *Average growth inhibition around agar wells of S. aureus for each treatment group. Error bar is one standard deviation.*

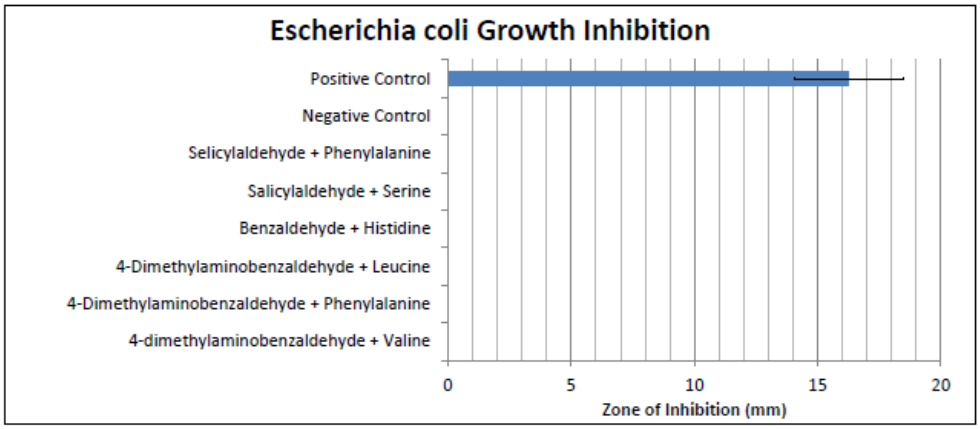

Figure 5. *Average growth inhibition around agar wells of E. coli for each treatment group. Error bar is one standard deviation.*

5. Conclusion

We have successfully synthesized and partially characterized Schiff base ligands derived from amino acids (serine, alanine, valine, histidine, phenyalanine and leucine). The melting/decomposition temperatures of the schiff base ligands are different from the amino acids from which they were derived, suggesting that new compounds were formed. This is further confirmed by the presence of the prominent imine bond (C=N) in the IR spectra of all the bases. The Schiff bases are coloured compounds as confirmed in literature and very soluble in water. This is an important feature in these compounds since they are intended to be used in biological systems. In addition, the schiff base ligands show no bacterial growth inhibitory properties or toxicity effects against gram positive and gram negative bacteria (*S. aureus and E. coli).* As further work, we will completely characterized the Schiff-bases and their metal complexes using ^{1}HNMR and ^{13}CNMR, X-Ray crystallography, biological activities, etc. The main aim of this paper is to synthesized and completely characterized their metal complexes with schiff-bases as ligands derived from amino acids.

Acknowledgements

We are highly indebted to the Department of Mathematical and Physical Sciences, Chemistry Research Laboratory, Concordia University of Edmonton, AB, Canada for

providing their laboratory space and chemicals used in this research.

References

[1] A. M. Abu-Dief, and I. M. A. Mohamed, Beni-Suef University Journal of Basic and Applied Sciences, 2015, (4), pp 119-133.

[2] a. H. L. Singh and J. Singh, Int. J. of Inorg. Chem., 2013, (vol 2013), pp 1-10, b. K. Ghosh et al., RSC Advances, 2018, (8), pp 28216-28237.

[3] a. M. Alias, H. Kassum and C. Shakir, JAAUBAS, 2014, (vol. 15), pp 28-34, b. Mohamed Gaber et al., Journal of Iranian Chemical Society, 2019, (16), pp 169-182.

[4] Kangah Niameke Jean-Baptiste et al., International Journal of Pharmaceutical Science Invention, 2019, (8), issue II, pp 48-54.

[5] N. K. Chaudhary and P. Mishra, bioinorganic chemistry and applications, 2017, pp 1-13.

[6] T. Mangamamba, M. C. Ganorkar, and G. Swarnabala, International Journal of Inorganic Chemistry, 2014, pp 1-22.

[7] Emad Yousif et al., Arabian Journal of Chemistry, 2017, (10), pp S1639-S1644.

[8] Majid Rezaeivala, Journal of Saudi Chemical Society, 2017, (21), pp 420-424.

[9] ElieneLeandrodeAraújo et al., International Journal of Biological Macromolecules, 2017, (95), pp 168-176.

[10] Rathore et al., Eur. J. Chem. 2010, pp. S566–S572 7 (S1).

[11] E. Yousif et al., Arabian, J. Chem., 2013, pp 1-5.

[12] Chohan Z. H., Arif M., Akhtar M. A., Supuran C. T., Bioinorganic Chemistry and Application, 2006, pp 1-13.

[13] El-Sherif A. A., Aljahdali M. S., Journal of Coordination Chemistry, 2013, 66 (19): pp 3423-3468.

[14] Rimbu C., Danac R., Pui A., Chem Pharm Bull., 2014, 62 (1), pp 12-15.

[15] De Souza A. O., Galetti F. C. S., Silva C. L., Bicalho B., Parma M. M., Fonseca S. F., Marsaioli A. J., Trindade A. C. L. B., Gil R. P. F., Bezerra F. S., Quimica Nova, 2007, (30), pp 1563-1566.

[16] Guo Z., Xing R., Liu S., Zhong Z., Ji X., Wang L., Li P. Carbohydr Res., 2007, (342), 1329-1332.

[17] G. L. Miessler, P. J. Fisher, and D. A. Tarr, Inorganic Chemistry, 2013, 5th Edition.