SYNTHESIS, SPECTRAL CHARACTERIZATION AND PRELIMINARY ANTIMICROBIAL SCREENING OF PHENYLALANINE AND ITS COPPER(II) AND COBALT(II) COMPLEXES

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Abstract:

Complexes of Copper(II) and Cobalt(II) derived from the ligand Phenylalanine were synthesized and characterized using UV-visible spectra, FT-IR spectroscopy, melting point and solubility studies. The results indicate Cu(II) was light pink, Co(II) deep blue and melted within the range of 76-170°C. The solubility test on the ligand and its metal(II) complexes revealed their solubility in ethanol, 1,2-propandiol, DMSO and chloroform. The FT-IR spectra revealed the presence of C=N, N-O, O-H, C-C, C=C, C=N, N-H, C-H, C=O, C-N and C-O which implies that the ligand is a bidentate ligand coordinating via the azomethine Nitrogen and the phenolic Oxygen. The ligand and complexes were evaluated for their antibacterial activity against Staphylococcus aureus, Streptococcus pyogenes, Salmonella typhi and Klebsiella pneumonia strains. The result of microbial screening showed that the complexes had moderate antimicrobial activity and the complexes were more active than the ligand.

Keywords: Antimicrobial, Coordination compounds, Ligand, Metal complex, Synthesis

Introduction

Much attention is being paid to coordination compounds in recent times as potential antimicrobial agent. This is due to the improved activity of drugs administered as complexes as there is much resistance to the already available antibiotics (Ama *et al*., 2017). It has also been reported that coordination compounds have wide application in food industry, dye industry, analytical chemistry, catalysis, fungicidal, agrochemical and biological activities (Zhai *et al*., 2015; Dudhat and Kurkarni, 2018; Salga *et al*., 2018). However, with growing incidences of infectious diseases there has been increasing emphasis on the screening of new and more effective antimicrobial drugs with low side effects (Acharjee *et al.*, 2015). Schiff base complexes are considered to be among the most important stereo chemical models in main group and transition metal coordination chemistry due to their preparative accessibility and structural variety. A considerable number of Schiff base complexes have potential biological interest, being used as more or less successful models of biological compounds. They have not only played a seminal role in the development of modern coordination chemistry, but also considered as key points in the development of inorganic biochemistry, catalysis and optical materials (Yiase *et al.*, 2014). It has been suggested that ligands with nitrogen and oxygen donor systems might inhibit enzyme production. This is because the enzymes which require these groups for their activity appear to be especially more susceptible to deactivation by the metal ion upon chelation (Selma and Sedat, 2014; Siddappa and Nabiya, 2015). Such compounds include coordination compounds of amino acids, such as phenylalanine. Phenylalanine and its metal complexes have been reported to be biologically active against a wide range of microorganisms (Fatih *et al*., 2018; Ozlen and Melike, 2020). In the present study, we have reported the synthesis, characterization and antimicrobial study of phenylalanine and its Co(II) and Cu(II) complexes.

Materials and Methods

Reagents/solvents and instrumentation

Chemicals and solvents were of analytical grade and used without further purification. The phenylalanine, copper(II) sulphate, and cobalt(II) sulphite, among other reagents were purchased from Coba Chemie Prt Ltd and Guang dong Guanghua Chemical Factory Co. Ltd.

The FTIR (Fourier Transform Infared Spectroscopy) of the desired compounds were carried out on a Thermo Scientific NICOLET iS5 FTIR spectrometer from 4000 - 400 cm^{-1} at 25°C using KBr plates.

Melting point (°C) were determined with aid of Electro-thermal 9100 melting point machine.

Preparation Of Co(Ii) Phenylalanine

The preparation of Co(II) Phenylalanine was carried out by using a method described by Faliah *et al* (2014) with slight modifications. Ethanol solution of phenylalanine (20 ml, 0.04 mole, 6.61 g) was gently poured into 20 ml (0.02 mole, 5.62 g) ethanol solution of Cobalt(II) sulphate placed on a heating mantle with constant stirring. The heating was constant at the temperature of 60°C with a continuous stirring using magnetic stirrer for about 10 mins until the appearance of a precipitate. The precipitate was left for about 1 hr to cool resulting to crystal formation. The crystal was collected by filtration, washed with cold ethanol and dried in a desiccator containing silica gel for 24 hrs (Faliah *et al*., 2014).

Preparation of Cu(II) Phenylalanine

The preparation of Cu(II) Phenylalanine was carried out by using a method described by Faliah *et al* (2014) with slight modifications. Ethanol solution of phenylalanine (20 ml, 0.04 mole, 6.61 g) was gently poured into 20 ml (0.02 mole, 4.99 g) ethanol solution of Copper(II) sulphate placed on a heating mantle with constant stirring. The heating was constant at the temperature of 60°C with a continuous stirring by a magnetic stirrer for about 10 mins. until the appearance of precipitate. The precipitate was left for about 1 hr to cool leading to crystal formation. The crystal was collected by filtration, and washed with cold ethanol, dried in a desiccator over a silica gel for 24 hrs (Faliah *et al*., 2014).

Antibacterial studies

The antibacterial activity of the ligand and its complexes was evaluated by the serial broth dilution method in a medium of agar nutrient on gram-positive bacteria (*Streptococcus pyogenes* and *Staphylococcus aureus*) and gram-negative bacteria (*Klebsiella pneumoniae* and *Salmonella typhi*) using nutrient agar.

Results and Discussion

Table 1 represent results for the synthesis of phenylalanine and its Co(II) and Cu(II) complexes which were light pink and deep blue respectively. The complexes were crystalline in nature and have high melting points. The Analytical and physical data of the ligand and complexes are shown in Table 1 below:

Table 1: Physicochemical data of ligand and complexes

Compound	Colour/molecular weight	Yield (%)	Nature of compound	M.P(°C)
Phen	White (165.19)	74.58	Powdery	76-80
[Co(phen)$_2$]	Light Pink (389)	89.45	Crystalline	100-113
[Cu(phen)$_2$]	Deep Blue (393.5)	82.24	Crystalline	114-170

Key: Phen = phenylalanine, M.P = melting point

Infrared spectra

The IR bands assignment of ligand and complexes are shown in Table 2. In the spectra of the ligand (Phenylalanine), absorption bands were observed at 1462 cm^{-1} and 1912 cm^{-1} and are assigned to nitro group and O-H group respectively (Salawu *et al*., 2015). Bands were also observed at 1462 cm^{-1} 1683 cm^{-1} and are due to C-C and C=C respectively. Bands are also present at 1582 cm^{-1}, 2923 cm^{-1}, 1683 cm^{-1} which were assigned to N-H, C-H, C=O respectively. This is in agreement with other works (Yiase *et al*., 2014; Jayaprakash *et al*., 2016).

For Cobalt(II) complex, the characteristic absorption band located at 1462 cm^{-1}, 1376 cm^{-1} is attributed to Nitro vibration. The band at 2923 cm^{-1} which is broad and weak is due to O-H in the ligand which indicate the involvement of oxygen as a lone pair donor during complexation from the carboxylic acid functional group in the ligand. There are also absorption bands at 749 cm^{-1} and 628 cm^{-1} attributed to M-O and M-N respectively. With this, it can be concluded that the ligand is a bidentate ligand coordinating via the nitrogen and the carboxylic oxygen.

For Copper(II) complex, the characteristic absorption occurred at 2923, 2853, 1458 and 1376 cm^{-1}. The broad absorption peak with high intensity is attributed to O-H. Thus, it can be concluded that the ligand is a bidentate ligand coordinating via the amine N and the phenolic or alcoholic oxygen (Jayaprakash *et al*., 2016). Bands were observed in the region of 737-739 cm^{-1} and 641 cm^{-1} which were assigned to M-O and M-N respectively.

In the work of Faliah *et al*., (2014), the infrared spectra of the synthesized compounds showed the presence of C=N stretching band at 1612 cm^{-1} for the first ligand; this band was shifted to lower frequency (1592 cm^{-1}) in the spectra of the chelate complexes while the C=N stretching band for the second ligand appeared at 1576 cm^{-1} and this band was shifted for the metal complexes to (1594-1580) cm^{-1} suggesting coordination of metal ion through nitrogen atom of azomethine group.

This work is in agreement with many works which reported a shift in the position of the bands upon complexation which has adverse effect on the properties of the complexes compared to the ligand (Fatih *et al*., 2018; Ozlen and Melike, 2020).

Table 2: FTIR bands of Ligand and Complexes

Compound	(N-O)	(O-H)	(C-C)	(C=C)	(C=N)	(N-H)	(C-H)	(C=O)	(C-N)	(C-O)	M-O	M-N
Phen	1462	1912	1462	1683	1683	1582	2923	1683	1038	1038	-	-
[Co(phen)$_2$]	1377	2923	1463	1634	1580	1633	2923	1738	1074	1074	749	628
[Cu(phen)$_2$]	1377	2923	1458	1623	1603	1623	2923	-	1092	1092	737	641

Key: Phen = Phenylalanine, M= metal (Co,Cu)

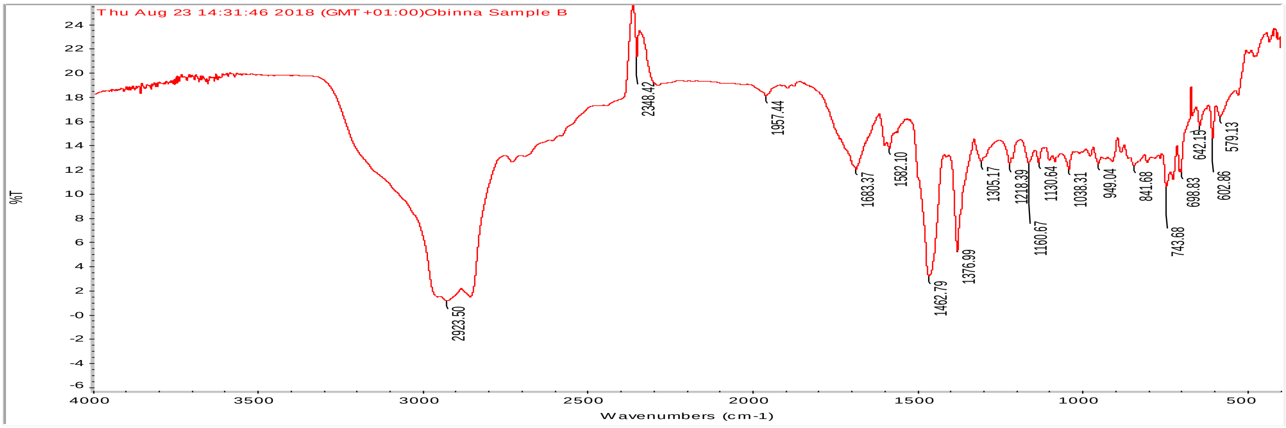

Figure 1: FT-IR Spectra of phenylalanine

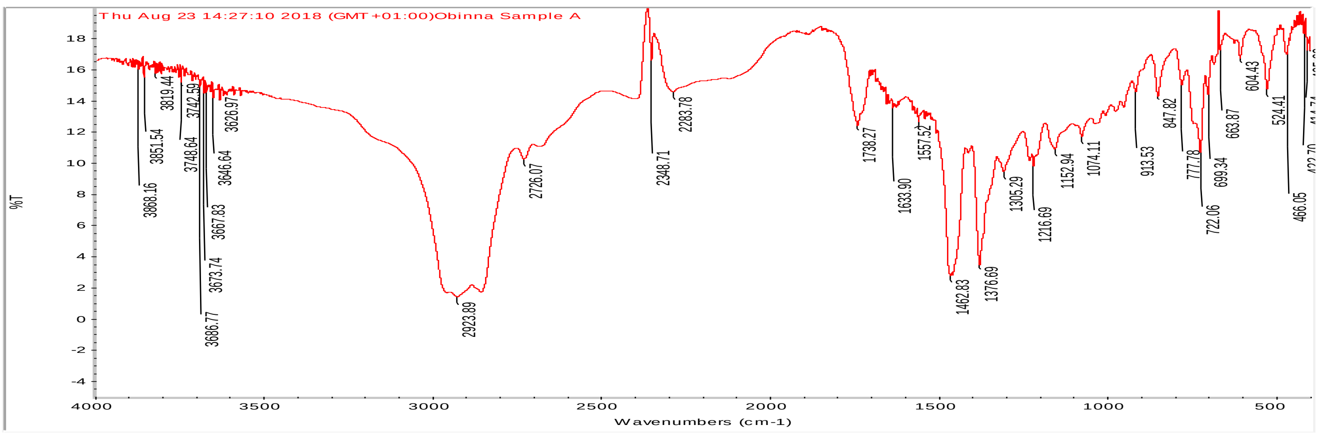

Figure 2: FT-IR Spectra of Cobalt(II) phenylalanine

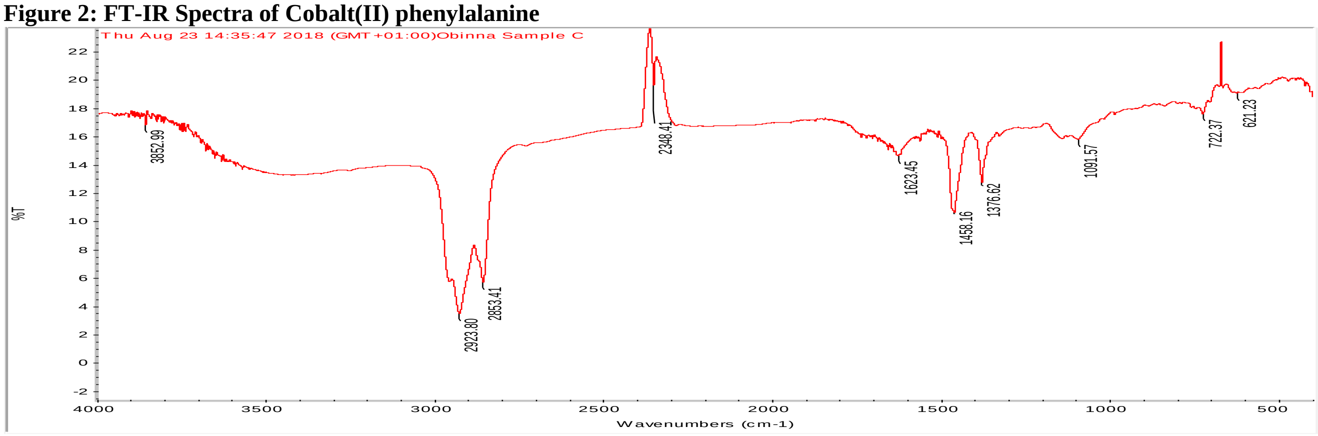

Figure 3: Spectra of Copper(II) phenylalanine

Antimicrobial activity

The *antimicrobial activity* of the investigated compounds was tested against microorganisms *Staphylococcus aurens, Streptococcus pyogenes, Salmonella typhi* and *Klebsiella pneumoniae*. The *growth inhibition* zones were measured and the results are summarized in Tables 3, 4 and 5.

Table 3: Antimicrobial Activities of Phenylalanine on Selected Micro organisms

		Zone Inhibition (mm)		
Conc. µg/ml	Sta	Str	Sal	Kle
100	4	4	2	2
200	4	4	1	0
400	4	2	1	0
800	3	2	0	0
1600	2	2	0	0
MIC	>3	>2	2	2

Table 4: Antimicrobial Activities of Cobalt(II) Phenylalanine on Selected Micro Organisms

		Zone Inhibition (mm)		
Conc. µg/ml	Sta	Str	Sal	Kle
100	4	4	2	2
200	4	4	1	0
400	4	4	1	0
800	3	2	0	0
1600	2	2	0	0
MIC	>3	>2	2	2

Table 5: Antimicrobial Activities of Copper Phenylalanine Complex on Selected Microorganisms

		Zone Inhibition (mm)		
Conc. µg/ml	Sta	Str	Sal	Kle
100	4	4	2	3
200	4	3	2	2
400	3	3	2	2
800	3	2	1	1
1600	3	1	1	1
MIC	3	3	2	2

The ligand showed low activity against *Streptococcus pyogenes and Klebsiella pneumoniae* at 100 µg/ml but showed moderate activity against *Staphylococcus aureus and Salmonella typhi* at all concentrations.

Cobalt(II)-Phenylalanine complex showed activity against *Staphylococcus aureus* and *Streptococcus pyogenes* at all concentration. There was no activity for *Salmonella typhi* at concentration of 800 µg/ml and 1600 µg/ml while for *Klebsiella pneumoniae*, the complex showed no activity at 200, 400, 800 and 1600 µg/ml.

Copper(II)-Phenylalanine complex showed activity against *Streptococcus pyogenes, Salmonella typhi* and *Klebsiella pneumoniae* at all concentration. Copper(II)-Phenylalanine complex also showed low activity against *Staphylococcus aureus* at concentration 100 µg/ml and 200 µg/ml.

The antimicrobial studies of ligand and metal complexes showed that the complexes have higher activity than the ligand, which can be attributed to the complexation of the metal ions with the donor atoms of the ligand (Aderoju *et al*., 2014; Jayaprakash *et al*., 2016; Ama *et al*., 2017]. Chelation also might have improved the lipophilic nature of complex, which therefore favors it access through the lipid bilayers of the cell membrane and restricts microorganism (More *et al.*, 2017; Shanty *et al.*, 2017).

Conclusion

Upon this study, it has been observed that Phenylalanine and its Co(II) and Cu(II) complexes are active against the tested microorganisms and the complexes are more active than the ligand against tested microorganisms.

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Conflict of Interest

The authors declare that there is no conflict of interest.

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