

The DNA cleavage and antimicrobial studies of Co(II), Ni(II), Cu(II) and Zn(II) complexes of 4-pyridinecarboxaldehyde with 4-aminopyridine

K. L. P. Sheeja Lovely¹, and M. Christudhas²

1, 2 (Department of Chemistry and Research Centre, N. M. Christian College, Marthandam, India)

Abstract: The Schiff base complexes of Co(II), Ni(II) Cu(II) and Zn(II) have been synthesized from 4- pyridine carboxaldehyde with 4-aminopyridine. They have been characterized by elemental analysis, molar conductance, molecular weight determination, IR, XRD and SEM. The metal in the Schiff base complex is hexa coordinated which binds through pyridine nitrogen, azomethine nitrogen and nitrate units of metal salt used in the preparation. The antimicrobial and cleavage activity have also been studied for the synthesized compounds. The Schiff base and its metal complexes show a good activity against the bacteria *Staphylococcus aureus*, *E.coli*, *Klebsiella*, *Pneumonia* and fungi like *Candida albicans*, *Apergillus niger* and *Pencillium sp*. The antimicrobial results also indicate that the metal complexes are better antimicrobial agents as compared to the Schiff base.

Key words: 4-aminopyridine, 4-pyridinecarboxaldehyde, Antimicrobial activity, Nuclease activity Spectral studies.

I. Introduction

Heterocyclic compounds are widely distributed in nature and essential to much biochemical Process. These compounds are worth attention because of their biological activities and clinical usage Schiff base ligand forms a stable complex with different transition metal ions and has been the subject for thorough investigation because of their extensive application in wide ranging areas from material science to biological sciences [1-3]. The literature reveals that Schiff base ligands are excellent coordinating ligands. During the past three decades considerable attention has been paid to the chemistry of the complexes of the Schiff base containing nitrogen and other donors. This may be attributed to their stability biological activity and potential applications in many fields such as oxidation catalysis [4-6] electrochemistry etc, making the behavior of Schiff base complexes with transition metals a worthy research topic. The coordination chemistry of amino acid-Schiff base ligands is also of considerable interest due to their biological importance [7-9]. Schiff base complexes of transition metals are often useful as chemical nucleases due to their structure and physico-chemical properties. The interaction of these complexes with DNA has gained much attention due to their possible applications as new therapeutic agents. Because ligands both influence the reactivity of transition metal complexes with DNA and can also be varied in an easily controlled fashion, their presence and manipulation greatly facilitate the interaction between the complexes and DNA [10-12]. Many potent antibacterial and antifungal Schiff base compounds of heterocyclic compounds were reported [13-15]. In this paper we describe the synthesis and characterization of Schiff base ligand prepared by condensation of 4-pyridinecarboxaldehyde with 4-aminopyridine and its complexes with Co(II), Ni(II), Cu(II) and Zn(II) ions. The metal in the Schiff base complex is hexa coordinated binds through pyridine nitrogen and azomethine nitrogen.

II. Experimental

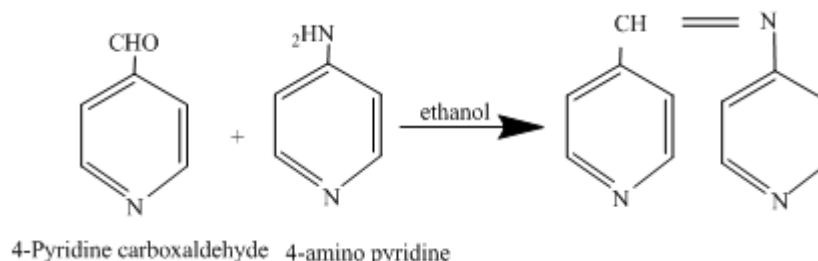
2.1 Materials and methods

All the chemicals and solvents used were of AR grade. All the reagents used for the preparation of the Schiff bases were obtained from Sigma Aldrich. Metal salts were purchased from Loba Chemie.

IR spectra of the ligand and their complexes have been recorded in KBr pellets at Shimadzu FT-IR 8201 spectrophotometer in 4000-200 cm⁻¹. Elemental analyses were performed with Perkin Elmer 240 analyzer. Molecular weight determination was done by Rast micro method using diphenyl as solvent. The XRD pattern of the complex was recorded on a Shimadzu XD-3 diffractometer using Cu-K_α-radiation ($\lambda = 1.54 \text{ \AA}$). The surface morphology of the complexes was studied using JSM-5610 scanning electron microscope.

2.2 Preparation of Schiff base ligand

The Schiff base ligand was prepared by mixing equimolar ethanolic solution of 4-amino pyridine with 4-pyridine carboxaldehyde in ethanol and refluxed for 1 hour, and then cool the mixture and the Schiff base was separated and recrystallized from ethanol and washed thoroughly with diethyl ether. (Scheme-1)



Scheme 1

2.3 Preparation of the solid complexes

Schiff base complexes were prepared by the addition of hot ethanolic solutions (60°C) of ligand and aqueous solution nitrates of Co(II), Ni(II), Cu(II) and Zn(II) in drop by drop in 2:1 molar ratio. The mixture was stirred for 8 hours at 60°C. The precipitated solids were washed thoroughly by ethanol and then by diethyl ether. The solid complexes were dried in vacuum desiccator.

2.4 Determination of antimicrobial activity

The in-vitro biological activity of the Schiff base and its metal complexes in DMSO were tested against the bacteria *Staphylococcus aureus*, *E. Coli*, *Klebsiella pneumonia* and the fungi *Candida albicans*, *Aspergillus niger*, *Penicillium. SP* by disc diffusion method using nutrient agar as medium and Amikacin, Flucanazole as control. The inhibition zone was developed at which the concentration was noted. The antimicrobial activity was estimated based on the size of inhibition zone in the discs [16-19].

III. Results and Discussion

All the metal complexes prepared above are coloured while Zn(II) complex is colourless and are stable towards air and have high melting point. The complexes are insoluble in water and common organic solvents but are soluble in DMF and DMSO.

3.1 Elemental Analysis

The analytical data suggest that all the complexes are mononuclear with a stoichiometric metal ligand ratio of 1:2. TABLE.1 indicates the physical characteristics and analytical data of complexes which are in good agreement with theoretical values.

Table.1 Physical characteristics and analytical data of ligand and its complexes

Compound	Colour Yield %	Empirical formula	Molecular weight	Melting point (°c)	Elemental Analysis calculated (found) %			
					C	H	N	M
C ₁₁ H ₉ N ₃ (L)	Pale yellow (80%)	C ₁₁ H ₉ N ₃	183 (179)	168-170	72.13 (71.86)	4.91 (4.13)	22.95 (21.97)	-
[Co(L) ₂](NO ₃) ₂	Brownish black (53%)	CoC ₂₂ H ₁₈ N ₈ O ₆	548.93 (541.51)	212-220	48.09 (48.98)	3.28 (3.79)	20.40 (19.56)	10.74 (11.64)
[Ni(L) ₂](NO ₃) ₂	Green (50%)	NiC ₂₂ H ₁₈ N ₈ O ₆	548.69 (536.23)	207-213	48.11 (48.23)	3.28 (3.92)	20.41 (20.84)	10.69 (9.13)
[Cu(L) ₂](NO ₃) ₂	Light green(70%)	CuC ₂₂ H ₁₈ N ₈ O ₆	553.55 (558.13)	220-225	47.69 (46.89)	3.25 (2.98)	20.23 (19.05)	11.48 (12.61)
[Zn(L) ₂](NO ₃) ₂	Grey white (65%)	ZnC ₂₂ H ₁₈ N ₈ O ₆	555.39 (546.45)	191-203	47.53 (48.88)	3.24 (2.99)	20.16 (19.19)	11.77 (12.96)

3.2 Magnetic susceptibility

The Magnetic susceptibility data obtained for the Schiff base complex systems are listed in TABLE. 2. The magnetic susceptibility values of Co(II) and Ni(II) complexes were found to be 5.0 BM, and 3.1 BM indicating an octahedral geometry around the metal ion [20-21]. The magnetic susceptibility value of Cu(II)

complex was found to be 1.8 BM indicates the octahedral nature of the complexes. The results also indicate the mononuclear and paramagnetic nature of Co(II), Ni(II), Cu(II) complexes, while Zn(II) complex was diamagnetic.

Table. 2 Magnetic Susceptibility data of the complexes

Complex	Magnetic susceptibility(BM)
[Co(L) ₂](NO ₃) ₂	5.0
[Ni(L) ₂](NO ₃) ₂	3.1
[Cu(L) ₂](NO ₃) ₂	1.8
[Zn(L) ₂](NO ₃) ₂	Diamagnetic

3.3 IR Spectra

The selected IR spectral data of the ligand and complexes are given in TABLE. 3 and the respective spectra are shown in Fig.1.1-1.5. In the IR spectrum of ligand, the absorption at 855 cm⁻¹, 1384 cm⁻¹ are due to the vibration of 4-substituted pyridine and C-H stretching vibration. The presence of -CH=N group, C-O group, and C-H group are identified by the presence of absorption at 1744 cm⁻¹, 2854 cm⁻¹ and 2925 cm⁻¹ respectively. The IR spectrum of the free ligand is compared with the spectra of metal complexes. The characteristic absorption bands 3451-3345 cm⁻¹ is assigned to -OH group of coordinated water. The absorption bands in the range 2864-2810 cm⁻¹ are due to the presence of C-H group. The band absorbed at 1457 cm⁻¹ is shifted to higher frequencies at 1643 cm⁻¹- 1676 cm⁻¹ in the complexes. This suggests that the nitrogens of pyridine ring are coordinated to the metal ion [22]. Another absorption bands in the range 781-720 cm⁻¹ is assigned to the (M-N bond) group with the central metal atom.

Table. 3 IR spectral data of ligand and its complexes

Ligand/Complex	$\nu_{\text{O-H(H}_2\text{O)}}$	$\nu_{\text{C-O}}$	$\nu_{\text{C-H}}$	$\nu_{\text{C=N}}$	$\nu_{\text{M-N}}$
C ₁₁ H ₉ N ₃ (L)	-	2854.99	2925.68	1744.43	-
[Co(L) ₂](NO ₃) ₂	3348.52	2810.11	2923.60	1660.63	781.36
[Ni(L) ₂](NO ₃) ₂	3386.38	2852.70	2924.61	1643.11	720.13
[Cu(L) ₂](NO ₃) ₂	3451.47	2864.12	2950.41	1676.51	770.54
[Zn(L) ₂](NO ₃) ₂	3345.72	2850.27	2972.46	1655.38	774.03

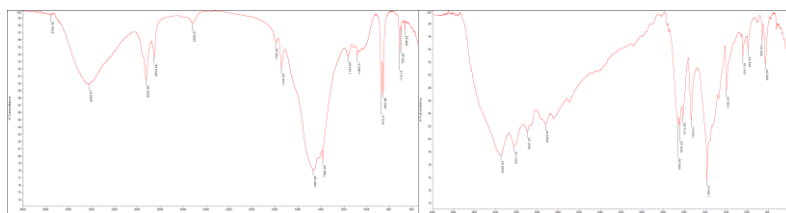


Fig.1.1 FT-IR spectrum of Schiff base ligand

Fig.1.2 FT-IR spectrum of Co(II) complex

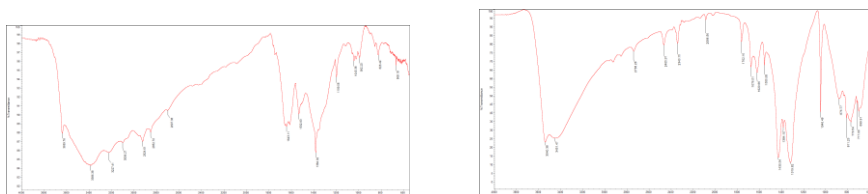


Fig.1.3 FT-IR spectrum of Ni(II) complex

Fig.1.4 FT-IR spectrum of Cu(II) complex

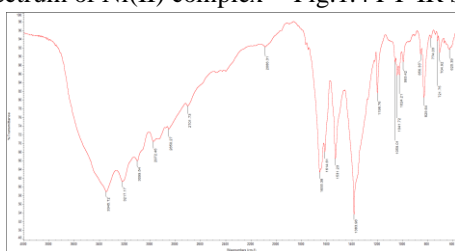


Fig.1.5 FT-IR spectrum of Zn(II) complex

Based on the above observations, the proposed structure of metal-Schiff base complexes $[M(L)_2] (NO_3)_2$ were given in Fig.1.6

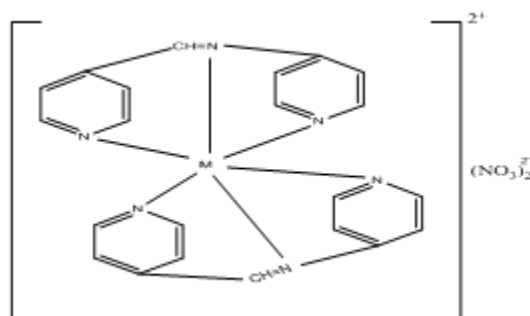


Fig.1.6 Proposed structure of Schiff base complexes of Co(II), Ni(II), Cu(II) and Zn(II) ions
M = Co(II), Ni(II), Cu(II) and Zn(II)

3.4 XRD Analysis

The powder XRD analysis of Ni(II) complex was carried out to find the crystalline or amorphous nature of the complex. The diffractogram was given in Fig.1.7. The strong and broad peak confirms the complex formation and the appearance of large feeble peaks indicates that the complex is microcrystalline [23]. The grain size of the complex is calculated using Scherer's formula. The calculated grain size 0.68 nm suggested that the complex is in microcrystalline state.

$$\text{Particle size (d)} = \frac{K\lambda}{\beta \cos \theta}$$

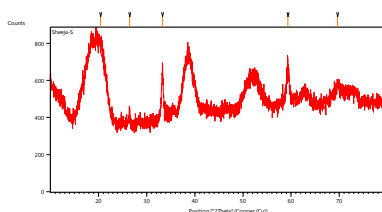


Fig.1.7 XRD Spectrum of Ni(II) complex

3.5 SEM Analysis

The surface morphology of Cu(II) complex was studied using SEM and the images were shown in Fig.1.8. The Cu(II) complex is microcrystalline in nature. The Cu(II) complex showed cocoon like appearance on higher magnification. Lower magnification showed grain like appearance.

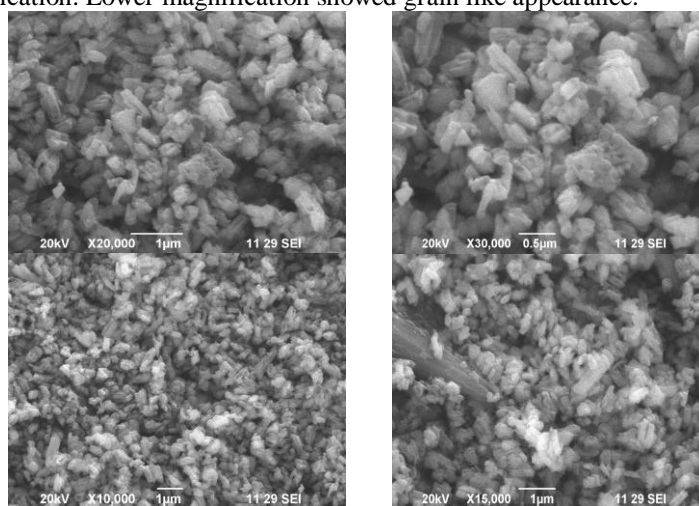


Fig.1.8 SEM images of Cu(II) complex

3.6. Biological screening

3.6.1 Antimicrobial Activity

The results of antimicrobial activity substantiate the findings of earlier researchers that biologically active compounds become active and less biologically active compounds become more active upon

coordination. Such enhancement in biological activity of metal complexes can be explained on the basis of Overtone's concept and chelation theory [24].

The present investigation suggest that all the metal complexes of the ligand bearing metal ion, pyridine ring, -N=CH- group, have comparatively more biological activity. This antibacterial activity serves as a basis for the chemical modification directed towards the development of new class of antimicrobial agents.

Table. 4 Antimicrobial activities of ligand and its complexes

Ligand/ complex	Antibacterial activity (mm)			Antifungal activity (mm)		
	<i>Staphylococcus aureus</i>	<i>E. Coli</i>	<i>Klebsiella pneumonia</i>	<i>Candida albicans</i>	<i>Aspergillus niger</i>	<i>Pencilium SP</i>
C ₁₁ H ₉ N ₃ (L)	8	7	8	7	6	6
[Co(L) ₂](NO ₃) ₂	12	11	10	8	9	9
[Ni(L) ₂](NO ₃) ₂	10	8	8	11	10	9
[Cu(L) ₂](NO ₃) ₂	9	8	9	8	7	8
[Zn(L) ₂](NO ₃) ₂	9	9	10	10	7	10
<i>Amikacin</i>	30	25	30	-	-	-
<i>Fluanazole</i>	-	-	-	20	30	25

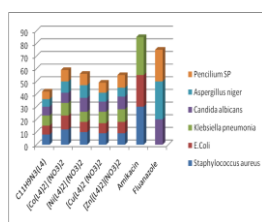


Fig.1.9 The variation of antimicrobial activity of ligand and its complexes

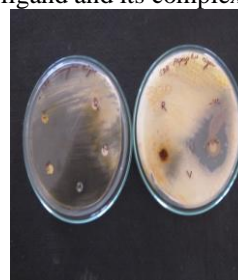
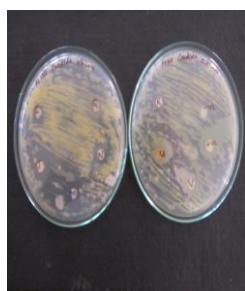


Fig.1.10 Images of antimicrobial activities of (inhibition zone) ligand and its complexes

3.6.2 Nuclease Activity

The nuclease activity of Co(II), Ni(II), Cu(II) and Zn(II) complexes were studied using gel electrophoresis and the respective photograph is shown in Fig.1.11. The cleavage efficiency of the complexes is compared with the control DNA to study the binding ability. The presence of smear in the gel diagram indicates the radical cleavage [25] by the abstraction of hydrogen from sugar units of DNA. The metal complexes were able to convert super coiled DNA into open circular DNA [26]. The reaction is modulated by the metallo complexes bound hydroxyl or peroxy radical generated from the oxidant H₂O₂. All the complexes cleave the DNA moderately.

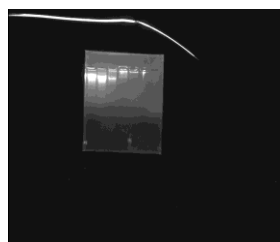


Fig.1.11 Gel diagram for M(II) complexes
M=Cu(II),Ni(II),Zn(II) and Co(II)

Lane 1-Control DNA

Lane4-DNA+Zn(II) complex

Lane 2-DNA treated with DMSO

Lane5-DNA+Ni(II) complex

Lane 3-DNA+Cu (II) complex

Lane6-DNA+Co(II) complex

4.5 Conclusion

The Schiff base ligand(L) was synthesized from 4-pyridine carboxaldehyde and 4-aminopyridine. They formed stable complexes (2:1) with transition metal ions such as Co(II), Ni(II), Cu(II) and Zn(II) in ethanol. The ligands and complexes are insoluble in water and common organic solvents, but are soluble in DMF and DMSO. The ligands and their complexes were characterized using spectral and analytical data. From the spectral and stoichiometric analysis, a hexa coordinated nature was assigned for the metal complexes, binds through two pyridine nitrogen atoms and one azomethine nitrogen. The nitrate group is present outside the coordination sphere. The XRD and SEM studies reveal that the complexes are micro crystalline. The antimicrobial study showed that Co(II), Ni(II), Cu(II) and Zn(II) complexes have moderately active against the tested organisms. Nuclease activity studies of complexes showed less cleavage.

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