INTERNATIONAL JOURNAL OF ADVANCE RESEARCH AND DEVELOPMENT

(Volume2, Issue2)

Available online at: www.ijarnd.com

Synthesis and Characterisation of new Heterocyclic Schiff base ligand derived from 4-Amino Antipyrine

E. Thamarai Selvi[1,2] , S. Mahalakshmi[2]

1 .Pachaiyappas College, Department of Chemistry, Chennai-30.

2. Anna Adarsh College for Women, Department of Chemistry, Chennai – 40

Abstract

A new heterocyclic Schiff base ligands and their complexes have been synthesized from 4-aminoantipyrine, thiophene-2-Carboxaldehyde and 2-amino benzoic acid. The ligands and its transition metal complexes of Cu (II), Co (II), Ni (II) and Zn (II) are characterized by UV, FTIR NMR and Mass studies. The antibacterial and antifungal activity of the ligands and the metal complexes were tested.

Key words: Heterocyclic Schiff base, 4-amino antipyrine, Transition metal complexes.

1. INTRODUCTION

Schiff bases derived from heterocyclic aldehydes has special centre of attraction in many areas like biological, clinical, medicinal, analytical and pharmacological field [1-3]. Among them 4-aminoantipyrine based heterocyclic's have gained great importance as it is abundant in nature and wide pharmacological activities [4]. 4-Aminoantipyrine is a pyrazole derivative which has antipyretic action [5]. It is used in the preparation of azo dyes [6]. 4-Aminoantipyrine is also used to protect against oxidative stress as well as prophylactic of certain diseases including cancer [7]. Several derivatives of antipyrine were also evaluated as analgesic [8], anti-inflammatory [9], antimicrobial [10] and anticancer activity [11-13]. Studies of a new kind of chemotherapeutic Schiff bases are now attracting the attention of biochemists [14-15]. Earlier work reported that some drugs showed increased activity when administered as metal complexes rather than as organic compounds [16-17].

The current work explains the synthesis of a new Schiff base ligands derived from 4-aminoantipyrine and its Cu, Co, Ni and Zinc complexes.

2. MATERIALS AND METHOD

All the reagents used for the synthesis 4-aminoantipyrine, 2-amino benzoic acid , metal perchlorates and Thiophene-2-carboxaldehyde were purchased from Sigma Aldrich and Himedia and were used as such. The solvents like ethanol, methanol, DMSO etc., were purified and dried by the standard procedures[18].The UV-Visible spectra were recorded on Schimadzu UV spectrometer in the wavelength range 200 – 800 nm.IR spectra were recorded on Schimadzu FTIR 8400S spectrometer in the wavelength range 4000-400 cm^{-1} using KBr pellet. The ^{1}H NMR spectra were obtained in $CDCl_3$ using Bruker, Ultra shield 3000 MKZ, Switzerland spectrophotometer.

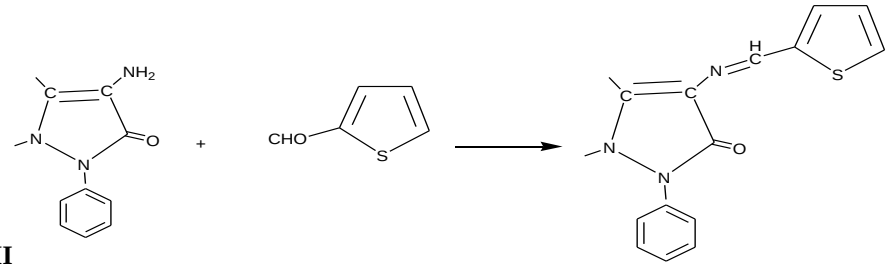
.

2.1. Synthesis of Schiff base ligands

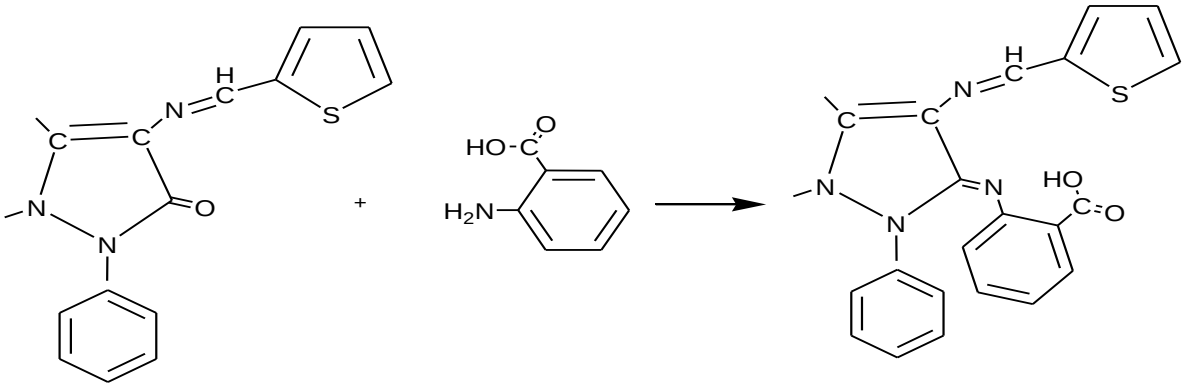
An ethanolic solution (1 mmol) of 4-aminoantipyrine was added to an ethanolic solution of Thiophene-2-carboxaldehyde (1 mmol) and the solution was refluxed for 3-4 hours with vigorous stirring and it was allowed to crystallize at room temperature. After 24 hours a shining yellow precipitate was obtained which was washed several times with ethanol and was dried at room temperature. The sample was recrystallised using hot ethanol. The solid (1 mmol) was added to an ethanolic solution of 2-amino benzoic acid (1 mmol). The mixture was

refluxed for about 36 hours. The completion of the reaction was followed using TLC. The brown solid product was separated, filtered and recrystallised from ethanol. The outline synthesis of ligands is shown in **Scheme 1**.

Step I



Step II



Scheme I. outline synthesis of Schiff base ligands

2.2. Synthesis of metal complexes

All the metal complexes were prepared following the general procedure. 1:1 combination of a solution of metal (II) perchlorates in ethanol (1 mmol) was refluxed with an ethanolic solution of the ligands (1 mmol) for approximately 5-6 hours. The solution was reduced to one-third of its volume on a water bath and was allowed to cool at room temperature. Microcrystalline precipitate was obtained after 2 days, which was then filtered, washed with ethanol and dried at room temperature.

2.3. Determination of antibacterial activity

The antibacterial activity was performed by disc diffusion method using Muller Hinton Agar medium [19, 20]. The bacterial species like Escherichia coli & Pseudomonas aeruginosa (gram positive) Staphylococcus aureus & Salmonella typhimurium (gram negative) were used in this study. Fresh bacterial cultures were used for this purpose.

The sample was weighed (10mg/10ml) and dissolved in 1% sterile DMSO to prepare appropriate dilution to get required concentrations (500, 250, 100 and 50 µg/ml) and were refrigerated. Standard solution as Gentamicin (100 µg/ml in 1% DMSO) used to compare the test solution. Whattman filter paper (No:1) was used to prepare discs approximately 6 mm in diameter, which are placed in hot air for sterilization After sterilization, the discs were loaded with different concentrations of prepared sample solutions and again kept under refrigeration for 24 hrs. Previously prepared paper discs were dispensed onto the surface of the inoculated MHA plates. Each disc was pressed down firmly to ensure complete contact with the agar surface. The discs were placed on the medium suitably apart. The plates were incubated at 5°C for 1 hr to permit good diffusion and then transferred to incubator at 37°C for 24 hrs.

2.4. Determination of antifungal activity

Candia albicans was selected as test organisms for the present study. The test fungi were procured from Microbial Type Culture Collection Centre, Institute of Microbial Technology Chandigarh. The organisms were maintained on Sabouraud dextrose agar.

Anti-fungal activity of the given samples was determined by disc diffusion method on SDA medium. The SDA medium is poured into the Petri plate. After the medium had been solidified the inoculums were

spread on the solid plates with sterile swab moisture with the fungal suspension. Using sterile forceps, the sterile filter papers (6 mm diameter) containing the sample 500, 250, 100 and 50 μg/ml were laid down on the surface of inoculated SDA plates. The plates were incubated at 27°C for 12 hrs and then plates were inverted and placed in an incubator set to respective temperature for 48 hrs.

2.5. Measurement of zone of inhibition

Then the microbial growth was determined by measuring the diameter of the zone of inhibition by using a dial caliper. The diameter which is less than 6 mm considered as not active against microorganism.

3. RESULTS AND DISCUSSION

3.1. IR spectra

The IR spectra of the ligands shows two peaks at 1632 and 1593 **ν cm^{-1}** which shows the formation of azomethine group (CH=N) .This has shifted to low frequencies in the metal complexes which confirms the involvement of azomethine N in bonding with the metal. Further it shows a peak at 759 **ν cm^{-1}** for the ligands which is due to the C-S-C linkage of thiophene group and this has shifted to lower frequencies in the metal complexes which show the involvement of sulphur group in complexation. Moreover the coordination of the ligands to the metal via carboxylic group can also observed from the difference of maxima positions for $\nu_{sym(COO-)}$ and $\nu_{asym(COO-)}$. The bands were noticed at 1388 and 1421 cm^{-1} for the free ligands. For the metal complexes $\nu_{sym\ (COO-)}$ and $\nu_{asym\ (COO-)}$ were observed at 1316-1320 and 1406-1405 cm^{-1}.The IR spectral data of the ligands and the complexes are given in **Table 1.**

Table 1. IR spectral data

Compound	**ν cm^{-1} (C=N)**	**ν cm^{-1} (COOH sym)**	**ν cm^{-1} (COOH asym)**	**ν cm^{-1} (C-S-C)**	**ν cm^{-1} (M-N)**	**ν cm^{-1} (M-O)**
Ligands	1632,1593	1388	1421	759	-	-
[Cu^{2+} L]	1613,1561	1391	1431	752	450	626
[Co^{2+} L]	1610,1542	1316	1406	758	468	629
[Ni^{2+} L]	1614,1548	1320	1406	758	482	625
[Zn^{2+} L]	1605,1551	1320	1405	750	470	628

3.2. UV – Visible spectra

The copper complex of ligands shows a broad band in the region 414 nm due to $^2B_{1g} \rightarrow {}^2A_{1g}$ transitions which favour the square-planar geometry around the central metal ion. The Co(II) complex exhibited a band at 504 nm which is assigned to $^1A_{1g} \rightarrow {}^1B_{1g}$ for square-planar geometry. Here, the nickel complex showed a band at 458 nm which is due to $^1A_{1g} \rightarrow {}^1B_{1g}$ for square planar geometry. The electronic spectral data of the ligands and the complexes were summarized in **Table 2.**

Table 2. Electronic spectral data

Compound	**Absorption Maximum nm**	**Tentative assignments**	**Geometry**
Ligand	241 350	INCT INCT	- -
Co(II) complex	240 354 504	INCT INCT $^1A_{1g} \rightarrow {}^1B_{1g}$ transition	- - Square planar
Ni(II) complex	250 319 458	INCT INCT $^1A_{1g} \rightarrow {}^1B_{1g}$ transition	- - Square planar
Cu(II) complex	305 323 414	INCT INCT $^2B_{1g} \rightarrow {}^2A_{1g}$ transition	- - Square planar

3.3. Mass spectra

The mass spectra of the ligands shows molecular ion peak at 416 m/z corresponding to $[C_{23}H_{20}N_4SO_2]$ ion whereas the copper and nickel complex shows the molecular ion peak at 479 and 481 m/z which confirms the stochiometry of the complex to be $[ML]ClO_4$ whereas M= Cu , Co, Ni, Zn.

3.4. $^1H_1$ NMR spectra

The $^1H_1$ NMR spectra of the Schiff base ligands and its complexes were recorded in $CDCl_3$. The Schiff base ligands shows a multiplet in the region 7.367- 7.550 δ, which is due to aromatic protons. The peak at 9.699 δ is attributable to azomethine protons and the shift in the spectrum of Zn complex suggests deshielding of >C ═ N group due to co-ordination with metal ion. The peak at 10.15 is due to the –COOH of 2-amino benzoic acid moiety present in Schiff base. The absence of this peak in zinc complex confirms the loss of –COOH proton during complexation.

Table 3 . $^1H_1$ NMR spectral data

Compound	**$\delta_{(N\text{-}CH3)}$**	**$\delta_{(C\text{-}CH3)}$**	**$\delta_{(CH=N)}$**	**$\delta_{(COOH)}$ of 2-amino benzoic acid**
Ligand	3.166	2.509	9.699	10.15
Zn(II) complex	3.2	2.51	10.01	-

3.5. Antimicrobial activity

Standard antibacterial agents were used for a comparative study. The synthesized ligands and its complexes were tested for their invitro antimicrobial activity. They were tested against the bacteria Escherichia coli & Pseudomonas aeruginosa (gram positive) Staphylococcus aureus & Salmonella typhimurium (gram negative) and fungi Candida albicans. The antibacterial and antifungal data were given in Table 4 , 5 & 6. From the results, it has been found that there is increased activity of the metal complexes compared to the ligands which is due to the lyophilic nature of the metal ion complexes [21, 22].

Table 4. Antibacterial activity (Gram positive)

compound	**Escherichia coli**				**Pseudomonas aeruginosa**			
	500 (µg/ml)	**250 (µg/ml)**	**100 (µg/ml)**	**50 (µg/ml)**	**500 (µg/ml)**	**250 (µg/ml)**	**100 (µg/ml)**	**50 (µg/ml)**
Ligand	13.34	11.70	10.20	7.00	12.98	11.45	9.73	8.27
Cu-complex	13.14	10.82	10.43	7.27	14.34	11.72	10.61	7.97
Co-complex	13.80	10.13	7.38	6.25	12.08	10.58	9.77	6.15
Ni-complex	11.03	8.55	8.14	6.69	12.48	10.45	8.86	7.10
Zn-complex	13.73	12.01	10.90	7.75	13.28	12.10	10.86	9.59

Table 5. Antibacterial activity (Gram negative)

compound	**Staphylococcus aureus**				**Salmonella typhimurium**			
	500 (µg/ml)	**250 (µg/ml)**	**100 (µg/ml)**	**50 (µg/ml)**	**500 (µg/ml)**	**250 (µg/ml)**	**100 (µg/ml)**	**50 (µg/ml)**
Ligand	-	-	-	-	8.57	7.02	6.65	6.15
Cu-complex	16.08	13.06	11.47	9.90	14.80	12.32	11.43	8.98

Co-complex	14.73	12.73	11.65	9.01	13.52	12.29	10.59	8.67
Ni-complex	8.90	7.58	7.05	6.10/	13.87	11.27	11.00	8.33
Zn-complex	14.13	11.00	10.64	8.66	14.35	11.95	11.24	10.28

Table 6. Antifungal activity

compound	**Candida albicans**			
	500 (µg/ml)	**250 (µg/ml)**	**100 (µg/ml)**	**50 (µg/ml)**
Ligand	10.22	7.546.92	6.92	6.17
Cu-complex	10.59	9.98	7.15	6.49
Co-complex	10.75	10.29	7.38	6.19
Ni-complex	10.77	10.50	9.31	7.87
Zn-complex	10.67	7.46	7.0	6.06

CONCLUSION

A new Schiff base macro cyclic ligands was synthesized and characterized by UV-Visible, IR, NMR and Mass spectra. The data of the complexes suggested a square planar geometry. Based on these data the proposed structure of the complex is shown in the figure.1 below. Evaluation of antibacterial and antifungal activity of the complexes against Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Salmonella typhimurium and Candida albicans exhibited that the complexes have potent biocidal activity than the ligands.

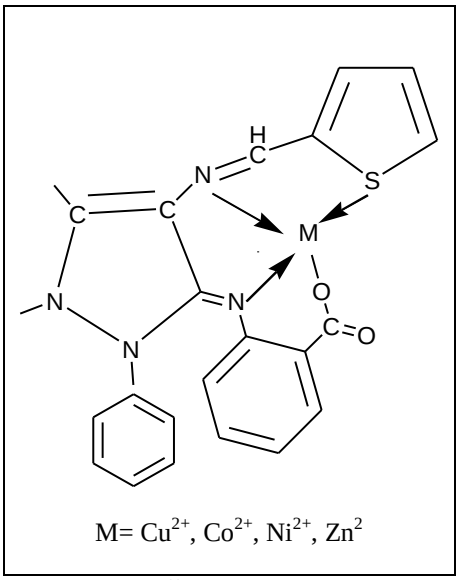


Fig 1.Schiff base metal complexes

ACKNOWLEDGEMENT

One of the authors (E. Thamarai Selvi) acknowledges the Head, Department of Chemistry and Principal, Pachaiyappas College for providing research facilities

6. REFERENCES

[1]. Sindhu KB, Raijuulal G, Mohanan K, Microwave Assisted Synthesis; Spectroscopic, Thermal and Biological studies of some Lanthanide(III) chloride Complexes with a heterocyclic Schiff base, Synth React Inorg Metal-Organic and Nano-Metal Chem, 39(1),2009, 24-30.

[2]. Thankamony M, Mohanan K, Synthesis, Spectral studies, Thermal Decomposition Kinetics, Reactivity and Antibacterial activity of some Lanthanide (III) Nitrate Complexes of 2-(N-indole-2-one)amino-3-carboxyethy-4,5,6,7-tetrahydrobenzo-thiophene, Indian JChem, 46A, 2007, 247-251.

[3]. Raman N, Dhaveethu Raja J, Sakthivel A, Synthesis, Spectral Characterization of Schiff base Transition Metal Complexes: DNA Cleavage and Antimicrobial Activity Studies, J Chemical Sciences, 119(4), 2007, 303-310.
[4]. Manjula B, Arul Antony S, Preparation, Characterization, Antimicrobial Activities & DNA Cleavage Studies of Schiff base Complexes Derived From 4-Aminoantipyrine, Asian Journal of Biochemical and Pharmaceutical Research, 3(1), 2013, 168-178.
[5]. Acheson RM, an Introduction to Heterocyclic Compounds, 3rd Ed., John Wiley & Sons, New York, 2009, 354.
[6]. Harikumaran Nair ML, Sheela A, Synthesis, Spectral, Thermal and Electrochemical Studies of Oxo Molybdenum(V) and dioxo Molybdenum(VI) Complexes of an azo dye derived from 4-amino-2, 3-dimethyl-1-phenyl pyrazol-5-one, Indian J Chem, 47A, 2008,1787-1792.
[7]. Teng Y, Liu R, Li C, Zhang H, Effect of 4-aminoantipyrine on Oxidative Stress Induced by Glutathione Depletion in single Human Erythrocytes using a Micro Fluidic device together with Fluorescence Imaging, J Hazardous Materials, 192, 2011, 1766-1771.
[8]. Turan-Zitouni G, Sivaci M, Kiliç FS, Erol K, Synthesis of some triazolyl-antipyrine derivatives and investigation of analgesic activity, Eur J Med Chem, 36(7-8), 2001, 685-689.
[9]. Lutsevich, AN, Bender KI, Reshetko OV, The Relationship Between Antipyrine kinetics, the Seromucoid content and the Xanthine Oxidase activity in the Plasma of rats with Acute and Chronic inflammation, Eksp Klin Farmakol, 58, 1995, 51-55.
[10]. Bondock S, Rabie R, Etman HA, Fadda AA, Synthesis And Antimicrobial Activity of Some New Heterocycles Incorporating Antipyrine Moiety, Eur J Med Chem, 43(10), 2008, 2122-2129.
[11]. Metwally MA, Gouda MA, Harmal AN, Khalil AM, Synthesis, Antitumor, Cytotoxic and Antioxidant Evaluation of some new Pyrazolotriazines attached to Antipyrine Moiety, Eur J Med Chem, 56, 2012, 254-262.
[12]. Kakiuchi Y, Sasaki N, Satoh-Masuoka M, Murofushi H, Murakami- Murofushi K, A Novel Pyrazolone, 4,4-dichloro-1-(2,4-dichlorophenyl)-3-methyl-5-pyrazolone, as a Potent Catalytic Inhibitor of Human Telomerase, Biochem Biophys Res Commun, 320, 2004, 1351-1358.
[13]. Sigroha S, Narasimhan B, Kumar P, Khatkar A, Ramasamy K, ManiV, Mishra RK, Abdul Majeed AB, Design, Dynthesis, Antimicrobial, Anticancer Evaluation, and QSAR studies of 4-(substitutedbenzylidene-amino)-1,5-dimethyl-2-phenyl-1,2-dihydropyrazol-3-ones, Med Chem Res, 21(11), 2012, 3863-3875.
[14]. Choi Y.K Chjo K.H Park SM and Doddapaneni N 1995 J.Electrochem Soc 142 4107
[15]. Katia B.Simon L Anne R, G rard C, Francoise D and Bernand M 1996 Inorg chem. 35, 387
[16].Hodnett EM and Dunn W.J 1972 J. med chem 15, 339
[17]. Hodnett EM and mooney P.D 1970 J.med chem. 13, 786 Raman.et.al 2007
[19].Awoyinka O, Balogun IO, Ogunnowo AA. (2007). Phytochemical screening and invitro bioactivity Of *Cnidoscolus aconitifolius* (Euphorbiaceae). *J Med Plant Res*; 1(3): 63-65.
[20].NCCLS. (1993) National Committee for Clinical Laboratory Standards. Performance standards fo antimicrobial disc susceptibility tests. *PA: NCCLS Publications* 2-5.
[21]. Raman.N, Raja .S.J and Sakthivel .A,J.Coord. chem.., 62, 691-709(2009)
[22]. Xu.S.P, Shi.L, Lv.P.C, Li.X.L and Zhu.H.L , J.Coord.Chem ., 62, 3198-3205(2009)

