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SYNTHESIS, SPECTRAL CHARACTERIZATION AND BIOLOGICAL STUDIES OF NOVEL SCHIFF BASE COMPLEXES DERIVED FROM 4,6-DIMETHYL-2-SULFANILAMIDOPYRIMIDINE AND 2-HYDROXY-3-METHOXYBENZALDEHYDE

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ABSTRACT

New Schiff base has been prepared from 4,6-Dimethyl-2-Sulfanilamidopyrimidine and 2-hydroxy-3-methoxy benzaldehyde in ethanolic media and then complexed with Mn(II), Co(II), Ni(II), Cu(II) and Zn(II) ions. The synthesized ligand and complexes were characterised by Elemental analysis, Molar conductance , Magnetic susceptibility, IR, UV, ^{1}H & ^{13}C NMR and EPR spectral studies. The reasonable shifts of FT-IR and NMR spectral signals of the complexes with respect to the free ligand confirm well coordination of Schiff-base ligand with the metal through imine nitrogen and oxygen atoms of Schiff base moiety. The Schiff base ligand and the complexes were screened for antimicrobial activity. From the analytical and spectral data, the stoichiometry has been found to be 1:2 for all the complexes. An octahedral structure has been proposed. All the new complexes were found to be active against bacteria and fungi.

KEYWORDS: **Schiff base,4,6-Dimethyl-2-Sulfanilamidopyrimidine, 2-hydroxy-3-methoxybenzaldehyde, Metal complexes, Biological activity.**

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INTRODUCTION

Sulpha drugs are chemotherapeutic agents whose molecular structures contain a 4-aminobenzene sulfonamide moiety. The antimicrobial activity of these drugs is believed from the structural resemblance between sulfanilamide group and p-amino benzoic acid where the sulpha drug mimics this metabolite and block folic acid synthesis in bacteria, there by causing cell death. Many sulpha drugs like sulphadiazine, sulphamethoxazole, sulphamerazine possess SO_2NH moiety as an important toxophoric function[1]. It has been reported that the biological activity has been increased when administered as metal complexes than as free ligands. The efficacy of the sulpha drugs can be enhanced upon co-ordination with a suitable metal ion[2]. Schiff base compound containing an imino group (-RC=N-) are usually formed by the condensation of primary amine with an active carbonyl group. Schiff bases are regarded as privileged ligands. Schiff base and their metal complexes are very popular due to their diverse chelating ability. They play an important role in both synthetic and structural research because of their preparative accessibility and structural diversity[3]. Metal complexes of Schiff bases are extensively studied due to synthetic flexibility, selectivity and sensitivity towards a variety of metal atoms. Schiff bases are used as pigments and dyes, catalysts, intermediates in organic synthesis and as polymer stabilizers. A number of Schiff's base molecules show biological activities including antibacterial, antifungal , antidiabetic, antitumour, antiproliferative, anticancer, anti-corrosion and anti-inflammatory activities[4-7].

The typical application of metal complexes of sulphadiazine has recently revived the usefulness of these compounds in medicine[8]. Indeed metal sulphadiazine complexes are now widely used to prevent bacterial infection during burn treatments. The interest in metal based sulfonamides was stimulated by the successful introduction and preparation of Ag(I) and Zn(II) sulphadiazine complexes to prevent various bacterial infections[9]. The present paper records the synthesis and characterization of Co(II), Ni(II), Cu(II),Mn(II) and Zn(II) complexes derived from 4,6-Dimethyl-2-Sulfanilamidopyrimidine and 2-hydroxy-3-methoxy benzaldehyde. The structures of the ligand and its metal complexes are characterized by elemental analysis, molar conductance and magnetic susceptibility measurements, IR, , UV, ^1H & ^{13}C-NMR and EPR. The biological activities are also studied against gram positive and gram negative bacterial and fungi organisms for Schiff base ligand and its complexes. The structure of Schiff base ligand confirmed in the present work is given in Figure 1

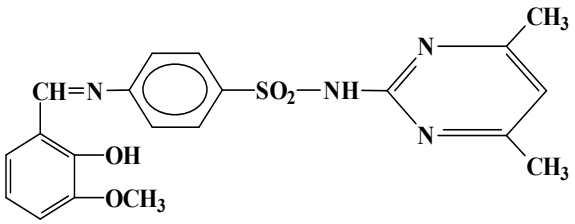


Figure 1
Structure of the ligand

EXPERIMENTAL

All the chemicals used were of analytical reagent grade (AR) and of highest purity available . Solvents were purified and dried according to the standard procedures. All metal (II) compounds were used as acetate salts. IR spectra of the complexes were recorded in KBr pellets with a Perkin Elmer RX1 FT-IR

Spectrophotometer in the 4000-400cm^{-1} range. The electronic spectra were recorded in DMF on a Perkin Elmer Lambda 35 spectrophotometer in the 190-1100 nm range. The ^{1}H& ^{13}C NMR spectra were recorded on a Bruker 400MHz FT-PMR spectrometer (DMSO-d_6). Elemental analysis of the ligand and complexes were obtained using Elementar Vario EL CHN rapid analyser.. The X-band EPR Spectrum were recorded on a Bruker ESP X-band EPR Spectrometer using powdered samples at a microwave frequency 9450MHz. Magnetic susceptibilities were measured on a Automagnetic Susceptibility meter (MSB-Auto) at room temperature. Melting points were determined using melting point apparatus (Elico) and are uncorrected. Conductivity measurements for the complexes were carried out on Elico Conductivity Bridge and a dip type conductivity cell using dimethyl formamide as solvent.

Synthesis of Schiff base ligand : (L) The Schiff base was prepared by the condensation of equimolar amounts of 4,6-Dimethyl-2-Sulfanilamidopyrimidine and 2-hydroxy-3-methoxy benzaldehyde in minimum quantity of ethanol. The resulting mixture was then refluxed on a water bath for 5 hours. A pale orange coloured solid mass separated out on cooling was filtered , washed and dried over anhydrous $CaCl_2$ in a desiccator. The purity of the ligand was checked by melting point, TLC and spectral data. The ligand is insoluble in some common organic solvents viz.acetone, benzene and soluble in polar solvents viz.DMF, DMSO.

Synthesis of metal complexes: Metal complexes were synthesized by mixing the hot solution of ligand (0.004 mole) in a minimum quantity of dimethyl formamide and ethanolic solution of metal acetates (0.002 mole). The resulting mixture was then refluxed in a water bath for 6 hours. The complexes obtained in each case were cooled, filtered and washed with ethanol several times to remove any excess of the ligand. Finally the complexes were washed with anhydrous diethylether and dried in a desiccator.

RESULTS AND DISCUSSION

The Schiff base ligand is synthesized by using equimolar quantities of 2-sulphanilamidopyrimidine and 2-hydroxy-3-methoxy benzaldehyde and is complexed with metal acetates according to the following equation

$$M(CH_3COO)_2.nH_2O + 2L \xrightarrow[\text{reflux}]{\text{ethanol\DMF}} ML_{2\text{-}2H}(H_2O)_2 + 2CH_3COOH + nH_2O$$

The metal complexes derived vary in their colour. All the complexes are stable, non-hygroscopic and coloured solids. The physical characteristics and micro analytical data of the ligand and metal complexes are given in Table 1 and Table 2

TABLE 1
Physical Characteristics of Schiff base ligand and their complexes

S. No	Ligand/ Complexes	Colour	Molecular Formula	M.P °C	Yield %	μ_{eff} (BM)	CN
1	L	Dark orange	$C_{20}H_{20}N_4O_4S$	105	70	-	-
2	$MnL_{2\text{-}2H}(H_2O)_2$	Brown	$C_{40}H_{40}N_8O_8S_2Mn$	135	55	5.65	6
3	$CoL_{2\text{-}2H}(H_2O)_2$	Brown	$C_{40}H_{40}N_8O_8S_2Co$	190	60	4.59	6
4	$NiL_{2\text{-}2H}(H_2O)_2$	Light green	$C_{40}H_{40}N_8O_8S_2Ni$	150	65	3.52	6
5	$CuL_{2\text{-}2H}(H_2O)_2$	Dark green	$C_{40}H_{40}N_8O_8S_2Cu$	165	70	1.99	6
6	$ZnL_{2\text{-}2H}(H_2O)_2$	Yellow	$C_{40}H_{40}N_8O_8S_2Zn$	170	65	dia	6

TABLE 2
Microanalytical data of Schiff base ligand and their complexes

S.No	Ligand/ Complexes	Elemental Analysis (%)(Calcd)found				%M (Calcd)found	Λ_m $ohm^{-1}m^2$ mol^{-1}
		C	H	N	S		
1	L	(58.25) 58.06	(4.85) 4.77	(13.49) 12.58	(7.76) 7.28	-	-
2	$MnL_{2\text{-}2H}(H_2O)_2$	(52.58) 52.50	(4.60) 4.55	(12.26) 12.20	(7.01) 7.00	(6.01) 5.99	15.65
3	$CoL_{2\text{-}2H}(H_2O)_2$	(52.34) 51.99	(4.58) 4.54	(12.21) 12.20	(6.97) 6.96	(6.42) 6.39	5.44
4	$NiL_{2\text{-}2H}(H_2O)_2$	(52.36) 52.30	(4.58) 4.57	(12.22) 12.20	(6.98) 6.96	(6.40) 6.30	7.02
5	$CuL_{2\text{-}2H}(H_2O)_2$	(52.09) 52.00	(4.55) 4.54	(12.15) 12.14	(6.94) 6.89	(6.89) 6.78	8.32
6	$ZnL_{2\text{-}2H}(H_2O)_2$	(51.98) 51.89	(4.55) 4.52	(12.13) 12.10	(6.93) 6.90	(7.08) 7.04	3.73

Molar Conductance and Magnetic Susceptibility Measurements

The observed molar conductances of all the complexes in 10^{-3}M DMF solution are found within the range of 3.73 - 15.65 $ohm^{-1}cm^2mol^{-1}$ showing their non-electrolytic nature. The magnetic data for Co ^{II} and Ni ^{II} complexes is consistent with octahedral geometry around the metal ion for both the complexes. The magnetic moment value of 1.99 BM for Cu (II) complex lies in the range expected for d^9 system which contains one unpaired electron with octahedral geometry[10] . Zn (II) complexes are found to be diamagnetic as expected. The observed magnetic moment value of 5.65 BM for the Mn (II) complex suggests the octahedral geometry.

Infrared Spectra

The infrared spectral data of the Schiff base and its metal complexes are recorded in Table 3. Schiff base showed a strong absorption band at 1600 cm^{-1} characteristic of v (C=N) whereas the broad band at 3448 cm^{-1} characteristic of hydrogen bonded v(O-H) stretching vibration [11]. The azomethine v(>C=N) band at 1600 cm^{-1} in Schiff base is shifted to higher frequency in Co(II), Mn(II),Ni(II),Cu(II), and Zn(II) by 27,25,14,54 and13cm^{-1} respectively indicates the co-ordination of azomethine nitrogen on complexation[12] . The shifting of phenolic (OH) at 3448 cm^{-1} in all the complexes suggests the coordination of phenolic oxygen after deprotonation[13]. The linkage with oxygen atom is further supported by the appearance of a band in the region around 466-476 cm^{-1} which may be assigned to v (M-O)[14]. A further evidence of the coordination of the N atom of the Schiff base with the metal atom is shown by the appearance of a new weak frequency band at 547-553cm^{-1} assigned to the metal nitrogen v(M-N)[15]. These new bands was observed only in the spectra of the metal complexes and not in Schiff base which confirmed the participation of the donor groups. The bands in the ligand due to v_{as} (SO_2) and v_s(SO_2) appear at 1155 cm^{-1} and 1332 cm^{-1} respectively. These bands almost remain unchanged in the complexes indicating that this –SO_2 group is not participating in coordination. This is confirmed by the unchanged v(S-N) and v(C-S) modes appearing around 975cm^{-1} and 869cm^{-1} respectively. The ring nitrogen (=N-) of the Schiff base does not take part in coordination, supported by unchanged band around 1261 cm^{-1}.

TABLE 3
IR and Electronic spectral data

Ligand/ Complexes	IR spectral data , cm^{-1}				Electronic spectral data, cm^{-1}
	υ(O-H)	υ(C=N)	υ(M-O)	υ(M-N)	
L	3448	1600	-	-	33579 ,28901
$MnL_{2\text{-}2H}(H_2O)_2$	3464	1627	547	470	35236
$CoL_{2\text{-}2H}(H_2O)_2$	3570	1625	547	472	38192,35961 , 34040
$NiL_{2\text{-}2H}(H_2O)_2$	3450	1614	549	466	35995
$CuL_{2\text{-}2H}(H_2O)_2$	3448	1654	553	476	35953
$ZnL_{2\text{-}2H}(H_2O)_2$	3452	1587	551	466	36249 , 35088

Electronic Spectra: Electronic spectrum of the ligand shows two high intensity bands at 33579 cm^{-1} and 28901cm^{-1} indicate $n \longrightarrow n^*$ and $\pi \longrightarrow \pi^*$ transitions respectively of the ligand moiety [16]. The electronic spectrum of the Mn(II) complex shows a band at 35236cm^{-1} is due to $^6A_{1g} \longrightarrow {}^4E_{g(D)}$[17]. The electronic spectra of Co(II) complex displays bands at 38192, 35961 and 34040 cm^{-1}. The first two bands corresponds to intra ligand transition of the organic moiety[18] and the latter corresponds to $^4T_{1g}$ (F) $\longrightarrow {}^4T_{1g}$(P) suggesting octahedral geometry of this complex. Ni(II) complex shows absorption band at 35995 cm^{-1} which is due to the $^3A_{2g} \longrightarrow {}^3T_{2g}$.The Cu(II) complex displays a band at 35953 cm^{-1} is attributed to $^2E_{(g)} \longrightarrow {}^2T_{2g}$[19]. Zn(II) complex displays high intensity bands at 36249 and 35088 cm^{-1}. This may be due to Ligand $\longrightarrow$ Metal charge transfer spectra[20]. The probable structure of complexes proposed in the present work is given in Figure 2.

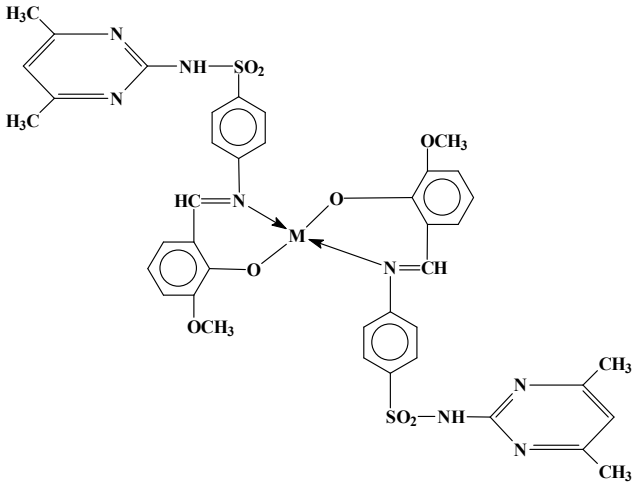


Figure 2
Structure of the Complex
M= Co(II), Mn(II), Ni(II), Cu(II) and Zn(II)

^{1}H NMR and ^{13}C NMR Spectra: The ^{1}H NMR Spectra of Schiff base and its complexes were recorded in DMSO (d_6). The azomethine proton (-CH=N-) in Schiff base appeared at δ = 8.92 ppm has been shifted to downfield in metal complexes. This confirms the coordination by azomethine nitrogen [21]. The aromatic protons in Schiff base appeared in the range at δ 6.60 to 8.09 ppm and metal complexes in the range δ 6.3 to 7.8 ppm . The disappearance of phenolic –OH proton signal at δ 12.77 ppm confirms the coordination by phenolic oxygen to metal ion[22]. The ^{13}C-nmr spectral data (imine at δ 165.0 ppm, aromatic C-OH at δ 123.83-127.85 ppm) for ligand supports the proposed structure.

EPR Spectra: The room temperature spectra of powdered samples were recorded at 9450MHz. Both parallel and perpendicular features of Cu are resolved in the spectra, which are characteristic of axial symmetry. The g value of Cu(II) complex is found to be around 2.2200 confirms the presence of unpaired electron in the dx^2-y^2 orbital of Cu(II).The g value are very close to those reported for a number of distorted Cu(II) complex. Moreover, the observed g value is less than 2.3, suggest the covalent nature of metal-ligand bonds in the complex. [23] The lines of this type usually observed are either due to the intermolecular spin exchange, which may broaden the lines or to the occupancy of the unpaired electron in the degenerate orbital. The nature and pattern of the EPR spectra (Figure 3) suggests an almost octahedral environment around the Cu(II) complex.

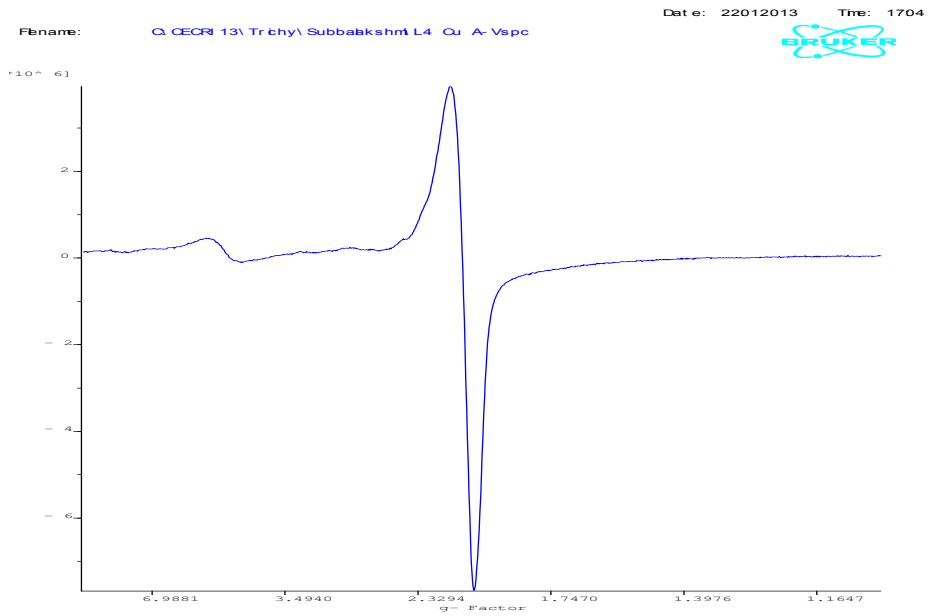


Figure 3
EPR Spectra for Cu(II)complex

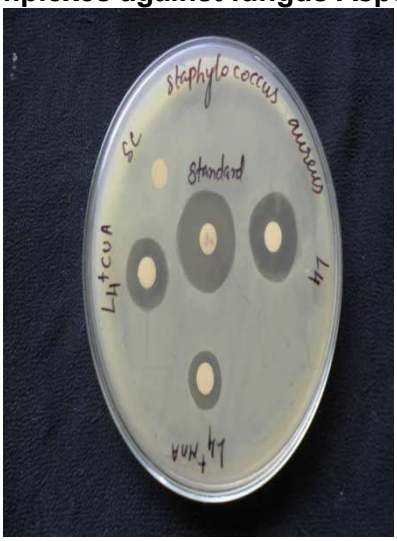
Antimicrobial activity: Antibacterial and antifungal activity of Schiff base ligand and its manganese cobalt, nickel,copper, and zinc complexes have been tested by disc diffusion technique[24,25]. The various gram positive and gram negative bacterial organisms such as Gram negative bacteria *pseudomonas aeruginosa, E.coli* Gram positive bacteria *staphylococcus aureus, Klebsiella aerogenes* and fungi *aspergillus niger* and *Mucor* were used to find out the antimicrobial activity. (Table 4). Filter paper discs of diameter 6mm were used and the diameters of zones of inhibition formed around each disc after incubating for a period of 72 hours at 25-30º C were recorded . Results were compared with standard drug Ciprofloxacin for bacteria and Nystatin for fungi at the same concentration. All the new complexes showed a

remarkable biological activity against bacteria and fungi (Figure4). From the results it is clear that the metal complexes are found to have more antimicrobial activity than the parent ligand.

a)Activity of ligand and Cu(II), Mn(II) complexes against fungus Mucor

b)Activity of Co(II),Ni(II) &Zn(II) complexes against fungus Aspergillus niger

c)Activity of ligand and Mn(II),Cu(II) against gram positive bacteria Klebsiella aerogenes



d)Activity of ligand and Mn(II),Cu(II) against gram positive Staphylococcus aureus

TABLE 4
Antimicrobial Activity of Schiff base ligand and complexes

Antimicrobial activity of the ligand and complexes	Staphylococcus aureus	Klebsiella aerogenes	E.coli	Pseudomonas aeruginosa	Mucor	Aspergillus niger
Ligand (L)	++	+++	+++	++	++	++
$[MnL_2(H_2O)_2]$	+++	+++	+++	+++	+++	+++
$[CoL_2(H_2O)_2]$	+++	+++	+++	++	+++	+++
$[NiL_2(H_2O)_2]$	+++	+++	+++	++	+++	+++
$[CuL_2(H_2O)_2]$	+++	+++	+++	++	++	+++
$[ZnL_2(H_2O)_2]$	+++	+++	+++	+++	++	+++

Standard= ciprofloxacin 5 g/ disc for bacteria ; Nystatin= 100 units/disc for fungi.
Highly active = +++ (inhibition zone > 15mm) ; Moderatively active = ++ (inhibition zone> 10mm) ;
slightly active = + (inhibition zone >5mm); Inactive = -- (inhibition zone <5mm)

CONCLUSION

The coordination ability of the newly synthesized Schiff base has been proved in complexation reaction with Mn(II), Co(II), Ni(II), Cu(II) and Zn(II) ions. IR, UV, ^{1}H NMR and magnetic measurements of the ligand and its complexes confirm the suggested coordination of the ligand through azomethine linkage. Based on these facts, an octahedral structure has been proposed for all complexes. The process of chelation dominantly affects the biological activity of the complexes that are potent against pathogens

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